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Environmental Assessment of the Alaskan Continental Shelf

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Final Reports of Principal Investigators
Volume 10. Biological Studies

U.S. DEPARTMENT OF COMMERCE
National Oceanic & Atmospheric Administration
Office of Marine Pollution Assessment

U.S. DEPARTMENT OF INTERIOR
Bureau of Land Management

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I

The facts, conclusions and issues appearing in these reports are based on interim results of an Alaskan environmental studies program managed by the Outer Continental Shelf Environmental Assessment Program (OCSEAP) of the National Oceanic and Atmospheric Administration (NOAA), U.S. Department of Commerce, and primarily funded by the Bureau of Land Management (BLM), U.S. Department of Interior, through interagency agreement.

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TABLE OF CONTENTS

Volume 10: Biological Studies

RU#	PI/Agency	Title	Page
019E	Warner, I.M. and P. Shafford -Alaska Dept. of Fish & Game, Kodiak, AK	Forage Fish Spawning Surveys - Southern Bering Sea	001
071	Costa, D.P. and G. L. Kooyman -University of California, La Jolla, CA	Effects of Oil Contamination in the Sea Otter, Enhydra Futris	065
078	O'Clair, E.C. et al. -Northwest & Alaska Fisheries Center, Auke Bay, AK	Reconnaissance of Intertidal Communities in the Eastern Bering Sea and the Effects of Ice - Scour on Community Structure	109

Project Completion Report
Research Unit No. 19 (Extension)
May 1977 - September 1977

Alaska Marine Environmental Assessment Project

FORAGE FISH SPAWNING SURVEYS - SOUTHERN BERING SEA

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TABLE OF CONTENTS

Page

Introduction 6

Acknowledgements 7

Current State of Knowledge 7

Study Area 11

Methods and Approaches for Data Collection 13

- Aerial Surveys 13
- Fish Sampling 16

Results 19

- Aerial Surveys 19
- Hydrography 27
- Fish Sampling 27

Discussion 44

- Fish Sampling 44
- Aerial Survey 52
- Hydrography 53

Conclusions 55

Appendices 57

Literature Cited 62

LIST OF TABLES

Page

Table 1 Entire coastline of the study area divided into census areas . . . 14

Table 2 Subjective index used for determining spawn intensity. 16

Table 3 Ageing methods used of forage fish 17

Table 4 Sub-areas, forage fish aerial surveys 21

Table 5 R.U. 19 aerial census results, OCSEAP forage fish aerial survey. . 21

Table 6 Mean herring fecundities, targets 1977 29

Table 7 Mean herring fecundities, Bristol Bay 29

Table 8 Miscellaneous species caught at Meshik/Port Heiden 1976 43

Table 9 Miscellaneous species caught at Meshik/Port Heiden 1977 44

LIST OF FIGURES

Page

Figure 1 The dock at Unalaska during height of herring run 9

Figure 2 Stroganof Point, north coast of Alaska Peninsula 12

Figure 3 Cape Kutuzof, north coast of Alaska Peninsula 12

Figure 4 Laboratory facilities used for forage fish at Port Heiden 18

Figure 5 Counting process of dried eulachon eggs for fecundity analysis . . 20

Figure 6 Mettler Analytical balance used for weighing dried fish eggs in fecundity processing 20

Figure 7 R.U. 19 (extension) 1977 study area . . . . [illegible]

Figure 8 Temperature and Salinity data obtained from three primary forage fish sampling stations 23

Figure 9 Weight frequency distribution of Pacific herring 24

Figure 10 Length frequency distribution of Pacific herring 25

Figure 11 Age group composition, herring 26

Figure 12 Three length/fecundity regression groups 28

Figure 13 Herring body length plotted against gonad weight, Togiak 31

Figure 14 Comparative condition factors of spring and fall Pacific herring 32

Figure 15 Capelin length distribution, Meshik/Port Heiden 33

Figure 16 Capelin weight distribution, Meshik/Port Heiden 33

Figure 17 Capelin age distribution as determined from otoliths, Meshik/Port Heiden 34

Figure 18 Age composition of capelin caught at Nunavarchak Bay, Togiak district 35

Figure 19 Length distribution of male-female capelin caught at Nunavarchak Bay, Togiak district 36

Figure 20 Relationship of length to fecundity of capelin from specimens taken at Nunavarchak Bay, Togiak district, June 1977 37

LIST OF FIGURES

Page

Figure 21 Size frequency composition of boreal smelt caught at Meshik 38

Figure 22 Weight frequency of boreal smelt caught at Meshik/Port Heiden during the spring-summer of 1977 field 38

Figure 23 Age composition of boreal smelt caught at Meshik/Port Heiden . . . 39

Figure 24 Incidence of various food items found in boreal smelt stomachs . . 40

Figure 25 Chart showing relationship of age and size of boreal smelt to number of circuli laid down before 1st annuli 41

Figure 26 Length, weight distribution of eulachon caught at Meshik/Port Heiden 42

Figure 27 Age composition, eulachon 42

INTRODUCTION

General Nature and Scope of Study

Forage fish research in the Bering Sea was initiated in fiscal year 1976 under the auspices of Research Unit 19 as a part of the Outer Continental Shelf Environmental Assessment Program (OCSEAP), of the National Oceanographic and Atmospheric Administration (NOAA). During that year field work began in May and continued until October. Field work consisted of aerial and ground truth investigations. Over a dozen temporary and permanent biological investigators were utilized by the Alaska Department of Fish and Game (ADF&G) to carry on all activities under the overall project guidance of the Principle Investigator, Mr. Louis Barton, ADF&G, Anchorage. His assistant, the senior author of this paper, supervised all field activities southwest of Smoky Point on the north coast of the Alaska Peninsula, while Mr. Barton was actual field supervisor for all activities north of Smoky Point.

Specific Objective

In the spring of 1977 OCSEAP funded the present study for the purposes of complementing the large bank of forage fish data accrued in 1976 under OCSEAP funding. The original purpose of this research unit was to study the nearshore spawning stocks of forage fish along the east coast of the Bering Sea in respect to relative abundance, spatial distribution, and basic life history facts concerning age and sexual maturity. This study's funding was at a minimal level to prevent complete loss of data continuity. Because of the limited funding, the nature of all 1977 activities was more austere than those completed in 1976, with the number of personnel reduced considerably. Since the basic life histories of all forage fish in the Bering Sea were either poorly known or not known at all, 1977 activities were invaluable in supplementing the knowledge of these species.

Relevance to Problems of Petroleum Development

It is known that surface borne pollutants are toxic to some species of forage fish. Kuhnhold (1970) studied the effects of certain crude oils upon Atlantic herring eggs, larvae and adults, and concluded that most crude oils were especially toxic to them in early life stages. Although it is not presently known how these types of pollutants would affect forage fishes in the Bering Sea, it is important to have an understanding of the temporal and spatial distribution and abundance of forage fish. If these factors are not known, long term effects of surface pollutants would be impossible to assess. If we know or have a fair assessment of where any species of forage fish is per time of year, the job of assessing the effects of petroleum related impacts would be greatly enhanced.

ACKNOWLEDGEMENTS

Many people have contributed to this study, including the following who assisted in project development and planning: ADF&G staff: Dennis Blankenbeckler, Glenn Davenport, Tim Jackinsky, Mike Jonrowe, Dorothy Lunsford, Jerry McCrary, and Ron Regnart. The following secretarial staff assisted in the production of reports: Kyle Watson, Beverly Burns, Elaine Damm, Lila Gowdy and Melayna McGuire. The following employees of the ADF&G collected the data: Brian Bue, Nell Fuqua, Dave Sczawinski, Mike Whelan and Dan Wiezorek. Jeff Collins of National Marine Fisheries Service (NMFS) in Kodiak provided the use of that agency's facilities and pilots Ed King and Howard McCubbin piloted most survey flights.

CURRENT STATE OF KNOWLEDGE

This study considered forage fish to be herring and sand lance or any form of the smelt family. All other fish are considered outside the scope of this report. Various cruises have recorded the presence of eulachon *Thaleichthys pacificus* and capelin *Mallotus villosus* in the Bering Sea - Bristol Bay area (Hart, 1973). Also boreal smelt* *Osmerus eperlanus* has been recorded as being present north to arctic Canada. Although the surf smelt *Hypomesus pretiosus* is recorded as ranging as far north as Chignik Lagoon, (Hart, 1973) it was found by the authors and also by investigators working in the vicinity of Izembek Lagoon (Smith, personal communication) to be present in the Bering Sea. The latter investigator found them in this region during 1972-73.

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*Confusion surrounds both the scientific and vernacular name of this fish. In this report, we shall refer to it in the vernacular as "boreal smelt" and scientifically as *Osmerus eperlanus*: numerous investigators refer to is as "toothed smelt", "rainbow smelt", "smelt" or "American smelt", the last being the least appropriate as the species is circumpolar in its distribution. Scientifically it is referred to as *Osmerus eperlanus* (McPhail and Lindsey, 1970).

Eskimos have known herring Clupea harengus pallasi to exist in the Bering Sea for thousands of years as they depend on them for food. Caucasians noted the occurrence of this fish when first arriving in the area, but they were first scientifically recorded by E. Nelson (1887) in his biological inventory work on the Bering Sea.

Substantial harvests of herring by the U. S. fishing industry took place during the early 1900's (Figure 1). Biological investigation of herring was initiated in 1929 when George Rounsefell completed his pioneer report on Pacific herring. Rounsefell worked on Pacific herring collected from the entire North American range of this fish, which are known to occur from San Diego Bay (possibly even Baja, California) northeast to Cape Bathurst in the Canadian arctic and northwest to the Russian arctic (Hart, 1973). Rounsefell employed the vertebral counts of herring from San Diego to Golovnin Bay in Norton Sound as a basis for population differentiation.

The next major work on Bering Sea herring was by Rumyantsev and Darda of the Soviet Union who conducted baseline investigations concerning the life history of Pacific herring in the Bering Sea during an inventory assessment project under the auspices of the Russian Fishing Agency, TINRO (Moiseev, P. A., editor, 1969). This work preceeded commercial entry by the U.S.S.R. into the herring fishery of this area. A compilation of domestic herring catch statistics was completed by the ADF&G (Randall, 1976) from the newly developed Togiak herring fishery.

In 1976, the ADF&G began investigations of Bering Sea herring and other forage fish under the OCSEAP R.U. 19 titled: HERRING SPAWNING SURVEYS, SOUTHERN BERING SEA. A project completion report was submitted in April of 1977 which included extensive work on herring (Barton, Warner, Shafford, 1977). This was the first extensive biological contribution to capelin in the Pacific basin, and a pioneer American effort into the life history of the herring in the Bering Sea.

Barton (1978) described further the biology of herring in the Norton Sound. In 1978 the North Pacific Fishery Management Council (NPFMC) funded a two year study conducted by the Alaska Department of Fish and Game. This study centered around herring and capelin, and further established life history parameters of both species concerning age, growth and spawning habitat demands (Barton, 1979). Also, a study pertaining to subsistence use of herring was funded by the NPFMC during 1978, and those findings incorporated into a project completion report by the Dames and Moore corporation that year (Hemming, 1978). Concern for the management of the foreign and domestic herring fishery in the Bering Sea caused NMFS to initiate efforts in this area. Wespestad (1977, 1978) compiled herring data from numerous sources with a view towards management information and consolidation of standing knowledge of herring in the Bering Sea. The preliminary Bering Sea management plan for herring in the study area contains a section concerning the biology and numerous other sections concerning herring (NPFMC, 1979).

It has been observed that herring overwinter in the deepwater areas northwest of the Pribilof Islands between latitudes 58 and 59 degrees north,

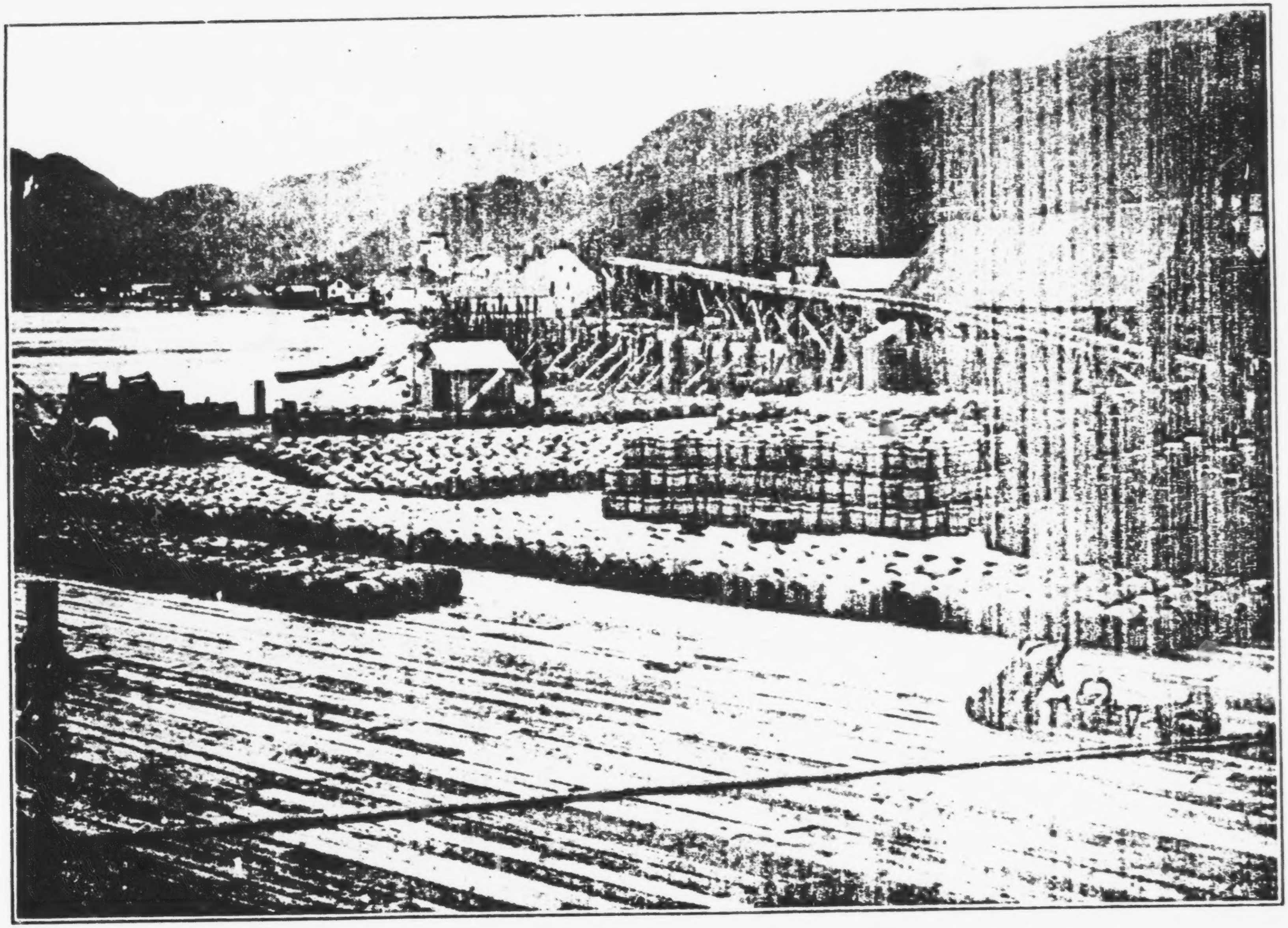

Figure 1. The dock at Unalaska during the height of the herring runs in August, 1928, showing piles of empty barrels and rows of packed barrels ready for shipment.

BEST DOCUMENT AVAILABLE

at depths of 105 to 137 meters, an area of approximately 2800 km (Favorite, et. al, 1977; Shaboneev, 1972). Japanese researchers have studied wintering herring in the area of 59°15'17" north (Takahashi, 1974) and presumably are familiar with their presence there.

The migration route of herring to their spawning grounds along the Bristol Bay-Bering Sea coast is hypothesized by Shaboneev (1972) to be two main routes passing north and south of the Pribilof Islands. How these routes were determined is not clear in this report, and the observation concerning the two routes towards the spawning grounds certainly needs further clarification. What is clear, however, is that most herring in the eastern Bering Sea spawn on the North American continent; Russian and Japanese scientists acknowledge this mutely, as they never consider that the massive body of herring wintering northwest of the Pribilof Islands go anywhere else to spawn.

Herring first arrive on the spawning grounds at varying intervals according to how far north those spawning grounds are (Warner, Barton, Shafford, 1977). Spawning runs in the Port Moller-Herendeen Bay complex occur in late May and early June, often extending into the middle of June. Spawning activity further north generally occurs later and some runs in the Bering Strait area occur as late as August.

The current state of knowledge concerning herring is greater than that of Osmeridae and Ammodytidae in the Bering Sea due to the lack of commercial interest in these fishes. Because of this there is no standing bank of knowledge for these groups outside of listing geographical occurrence. The authors consulted literature outside of the study area in order to develop methodology and gather life history data concerning the smelt family. The most important work concerning capelin was the exhaustive work completed by Templeman (1948), which pertains to capelin on the east coast of Canada and establishes many techniques for investigation of life history parameters.

No single comprehensive work exists concerning boreal smelt, yet McKensie's 1948 paper on the growth and age of boreal smelt in New Brunswick provided valuable insight into the problems of investigating this species. Other works on boreal smelt were reviewed, but most were too specialized or generalized for our purposes.

A great lack of prior information regarding eulachon* was encountered as there are no major works on this species outside of Smith and Saalfield's brief 1955 paper on the Columbia River Smelt. During the past two years, the authors have found only five papers dealing specifically with eulachon.

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*The common names as applied to eulachon are extremely chaotic, a condition which plagues all members of the smelt family. Eulachon are also called Columbia River Smelt, hooligan, candlefish and just plain smelt. Eulachon is a corruption of the French word for this fish which occurred in the Northwestern Chinook jargon, a common trade language of this area for over 125 years. In Alaska, all names are used in different combinations for all the smelts, and even for fish outside the smelt family. This condition makes anecdotal investigations concerning any of the smelt a matter of confusion.

A brief life history review will introduce each discussion section. This will serve to inform the reader immediately prior to an in depth species discussion.

STUDY AREA

The study area includes all coastal waters of the north Alaska Peninsula and Bristol Bay from Unimak Pass north to the Yukon River Delta including Nunivak Island, a total of 3,594 kilometers or 2,233 statute miles. Zoogeographically, this area includes sub-polar and taiga ecosystems typified by a continental shelf zone that is at least partially ice covered during all or some of the winter. The area contains some of the world's largest and most valuable fisheries partially or completely within its bounds. Perhaps the most dynamic feature of the study area is the Bristol Bay region which for 70 years has been the location of the largest sockeye salmon fishery in the world. Fish are of immediate importance to every living thing within this system, either directly or indirectly. The majority of the waters in the Bristol Bay region are turbid, a condition which precludes aerial spotting of fish schools. Weather conditions are mercurial, and human lives are lost each year due to this. The coast line within the bay itself is typified by low bluffs topped with tundra to the edges, which fall away steeply to expansive beaches which are open and exposed, or by low lying coasts littered with hundreds of ponds and wide, almost level beaches.

Outside of Bristol Bay to the north and west, geographic features change somewhat: in the Cape Newenham region to the north, sheer cliffs may tower hundreds of feet above the shoreline. To the southwest along the north coast of the Alaska Peninsula between Cape Sarichef and Cape Greig open beaches stretch for miles without indentations or coves interrupting the straightness of the coastline (Figures 2 and 3). Along this section of the study area are only four major bay systems: Port Heiden, Port Moller, Izembek Lagoon and Bechevin Bay. When combined, the shorelines of these systems total 665 kilometers, compared with 502 kilometers of open beaches along the Alaska Peninsula northeast to Cape Greig. This ratio of open to closed coastline is far different than most of Alaska's coasts. On its south coast, this portion of the Alaska Peninsula is mountainous becoming flat and extending north to the Bering Sea in a long gentle escarpment. The mountains, some actively volcanic, are capped with snow late into the summer, and the narrow passes in these ranges funnel southerly winds in such a fashion as to create dangerous and unpredictable winds on either coast.

It is over this 2,200 mile section of coastline that the North American continent joins the Bering Sea, and it was this area that the research responsibilities for this project existed. Field logistics for crews and aerial surveys were difficult being almost entirely dependent on air taxi services, which were frequently overbooked during the spring-summer months when project activity was at its peak.

Figure 2. Stroganof Point, north coast of the Alaskan Peninsula, looking toward Port Heiden, Mishik Mountains in the background, July 1977.

Figure 3. Cape Kutuzof, north coast Alaskan Peninsula, June 1976.

METHODS AND APPROACHES FOR DATA COLLECTION

The two primary approaches used to accomplish the objectives of this research unit were: 1) Aerial surveillance of spawning and/or moving schools of forage fishes and 2) operation of ground truth stations to collect specimens in order to determine species composition, life history parameters and age class composition.

Aerial Surveillance

The entire coastline of the study area was divided into census areas (Table 1). These divisions were made to facilitate digitizing data. The purpose of the surveys was to monitor the location, timing and relative abundance of spawning populations of herring and other forage fishes.

Single engine aircraft chartered from local air taxi operators were used for the most part to conduct aerial surveys. Survey data were recorded on aerial survey forms which were compatible with the OCSEAP file type 057 format. A survey was defined as a single flight on a single calendar day per observer(s). All weather conditions and chronological information were recorded for each census area surveyed along with geomorphic beach types. Weather data were usually determined visually from the aircraft's instruments, and by calling into the closest FAA flight information center. If instrumentation wasn't available, the observer estimated the values of wind direction by direct observation. Geological information was also gathered by direct observation. Water turbidity was recorded, and an overall value was arbitrarily assigned to general survey conditions encountered by each observer. Fish schools observed were enumerated separately unless they were within very close proximity of each other (within 300 meters), in which event each aggregation of schools was counted as one school. In this case, the number of schools in each aggregation were recorded, hence total individual schools seen per survey is recoverable. The estimated distance of each school from shore was recorded along with a subjective value of school size (large, medium, small). The location and species of each school were recorded along with the time it was observed.

Survey observations for each census were often peculiar to the observer. The favored altitude from which surveys were conducted differs amongst aerial forage fish spotters. The senior author favored 330 meters, whereas other observers preferred a somewhat lower or higher altitude. Generally though, censuses were conducted from an altitude of 300 meters at a speed of 220 kilometers per hour. Meterological conditions often required a survey altitude of less than 300 meters, and surveys frequently had to be terminated due to inclement weather. Polarized sunglasses were worn by observers to reduce surface glare, hence increase observation potential. Most OCSEAP personnel recorded their aerial data onto tape recorders in the aircraft for later transcription onto aerial survey forms.

Table 1. Entire coastline of the study area divided into census areas, a description of those areas including linear distance and mid-point. OCSEAP Forage Fish Studies, 1977.

Census Areas				Linear Shoreline Distance		Census Area 1/ Mid point (km)
Area	No.	North* Latitude	West* Longitude	Miles	km	
C. Sarichef-C. Mordivinof	1-A	54°38'30"	164°40'32"	18.0	29	14.5
C. Mordivinof-C. Lapin	1-B	54°54'20"	164°17'32"	13.7	22	11.0
C. Lapin-Chunak Point	1-C	55°03'15"	163°47'25"	31.1	50	25.0
Chunak Point-False Pass	2-A	54°58'28"	163°26'53"	20.5	33	16.5
False Pass-C. Krenitizin	2-B	54°57'28"	163°15'50"	46.0	74	37.0
C. Krenitizin-C. Glazenap	2-C	55°08'55"	163°14'08"	21.1	34	17.0
C. Glazenap-Moffet Pt. & Amak Is.	2-D	55°21'01"	162°41'30"	73.3	118	59.0
Moffet Pt.-Lagoon Pt.	3-A	55°48'21"	161°57'30"	70.8	114	57.0
Nelson Lagoon & Walrus & Kritskoi Is.	3-B	55°58'00"	161°07'40"	54.1	87	43.5
Herendeen Bay	3-C	55°42'50"	160°42'25"	75.2	121	60.5
Pt. Divide-Entrance Pt.	3-D	55°46'21"	160°46'16"	64.6	104	52.0
Entrance Pt.-Ilnik	4-A	56°20'30"	160°14'53"	62.1	100	50.0
Ilnik-Reindeer Creek	4-B	56°50'45"	158°57'29"	96.3	155	77.5
Reindeer Creek-Cinder River Lagoon	5-A	57°13'11"	158°20'05"	32.9	53	26.5
Cinder River Lagoon-Smokey Point	5-B	57°30'54"	157°50'59"	44.7	72	36.0
Smokey Pt.-Kvichak River	6	58°14'00"	157°30'00"	118.1	190.0	95.0
Kvichak River-Nusagak	7	58°38'50"	158°00'20"	79.5	128.0	64.0
Coffee Pt.-Tvativak Bay	8	58°23'10"	158°43'00"	83.9	135.0	67.5
Kulukak Bay	9	58°55'10"	159°36'30'	28.0	45.0	22.5
Metervik Bay-Togiak	10	58°52'00"	160°00'00"	46.6	75.0	37.5
Summit Island	11	58°51'00"	160°14'20"	6.4	10.3	Perimeter
Round Island	12	58°36'20"	159°58'00"	4.7	7.5	Perimeter
Crooked Island	13	58°43'40"	160°17'40"	14.9	24.0	Perimeter
High Island	14	58°45'00"	160°24'10"	10.9	17.5	Perimeter
Togiak-Asigyukpak Spit	15	58°48'30"	160°49'20"	77.7	125.0	62.5
Hagemeister Island	16	58°46'50"	160°42'00"	66.8	107.5	Perimeter
Asigyukpak Spit-Cp. Newenham	17	58°36'30"	161°43'20"	60.6	97.5	48.5
Cp. Newenham-Chagvan Bay	18	58°39'40"	161°52'00"	32.6	52.5	26.3
Chagvan Bay	19	58°50'00"	161°46'00"	14.3	23.0	11.5
Chagvan Bay-Goodnews Bay	20-A	58°54'40"	161°47'10"	20.2	32.5	16.3
Goodnews Bay	20-B	59°07'00"	161°36'10"	30.8	49.5	24.8
Goodnews Bay-Kuskokwin River	21	59°25'30"	161°51'20"	104.9	168.6	84.4
Kuskokwim River-Kolavinarak River	22	59°47'10"	163°46'20"	109.4	176.0	88.0

1/ Linear shoreline distance on either side of census area mid point.

* Latitude and Longitude of mid point of census area.

Table 1. (Cont'd.)

Census Areas				Linear Shoreline Distance		Census Area 1/ Mid point (km)
Area	No.	North* Latitude	West* Longitude	Miles	km	
Kolavinarak River - Cape Vancouver	23	60°29'00"	165°00'30"	47.7	76.8	38.4
Cape Vancouver - Minglick River	24	60°39'00"	165°09'00"	27.3	44.0	22.0
Nunivak Island	25	60°17'30"	165°41'00"	290.0	466.0	Perimeter
Hazen Bay	26	61°40'00"	165°00'00"	50.0	80.5	40.3
Angyoyaravak Bay	27	61°12'40"	165°33'30"	40.4	65.0	32.5
Hooper Bay	28	61°31'30"	166°02'00"	46.6	75.0	37.5
Kokechik Bay	29	61°39'40"	165°48'00"	35.7	57.5	28.8
Scammon Bay-Black River	30	61°57'00"	165°43'20"	60.6	97.5	48.8
Black River - North Mouth Yukon River	31	63°08'30"	164°33'50"	120.0	193.0	96.5

1/ Linear shoreline distance on either side of census area mid point.
* Latitude and Longitude of mid point of census area.

Fish Sampling

A series of sampling stations were established along the trackline of the aerial survey census flights. The location of these stations changed in reference to major spawning locations seen from the air by the project leader. A sampling crew was dispatched immediately to such locations if possible. Major spawning areas were examined by boat and foot surveys.

Test fishing was conducted at Meshik/Port Heiden continuously between May 21, 1977 and July 4, 1977. A short sampling period was conducted in the fall of 1977 between October 18 and October 19.

Beaches were walked at low tide and observations made as to egg density and distribution and types of spawning substrates utilized. Vegetation was identified by the use of Robert F. Scagel's Guide to Common Seaweeds of British Columbia, (1967). The density of spawn was recorded based on a subjective scale of deposition (Table 2).

Table 2. Subjective index used for determining spawn intensity for herring and capelin, Bering Sea OCS Forage Fish, 1977.

Spawn Intensity	
1. Very Light	1-2 layers
2. Light	3-4 layers
3. Medium	5-8 layers
4. Heavy	9-15 layers
5. Very Heavy	7-16 layers
6. Very Light	<20 eggs per cubic .25 meter
7. Light	20-40 eggs per cubic .25 meter
8. Medium	41-60 eggs per cubic .25 meter
9. Heavy	>60 eggs per cubic .25 meter

All test fishing at sampling stations was done with variable mesh gill nets ranging from about 28 to 80 mm (stretch mesh). These gillnets ranged from approximately 17 to 21 meters in length and 1.75 meters in depth and were fished by drift netting offshore or set netting onshore. Set netting was most commonly done by setting and picking the net on successive low tides and allowing it to fish when inundated by the high tide. Herring samples from offshore areas were made available to this investigator in 1976 through the cooperation of fisheries scientists on board the M/V PAT SAN MARIE, under charter to NMFS, Seattle. These forage fish specimens were taken incidentally to demersal and shellfish sampling in the Bering Sea. A few onshore samples were also obtained by the use of 45 meter hand beach seine, tapered from .7 meter to 1.3 meters in depth with a bag. Spawning capelin were obtained directly from the surf by hand.

Hydrological data was collected by the use of hand held thermometers calibrated to the nearest half degree Celsius. Salinity was determined at some stations by the use of NaCl hydrometer.

All fish specimens were measured and weighed to the nearest millimeter and gram respectively. Gonads were weighed to a tenth of a gram. When subsampling was necessary, variable mesh gill net catches were subsampled following specific instructions which insured that the subsample would represent the catch from each "panel" of the gill net.

Standard lengths were taken on herring (Eddy, 1969) and fork lengths were taken on all other species. All fish body weights were taken with a triple beam balance. Condition factors (K values) were determined by using formulae derived by Ricker (1962).**

In the case of species where ageing methods were relatively untried, ages were determined by a combination of methods. In the case of a species where age methods were firmly established, a single method was employed. Table 3 shows what age method was used on each species. Osmerids were aged using both otolith and scale. Methods in processing each type of bony part varied but essentially otoliths were processed by clearing off surface material by soaking them in a mixture of the enzyme papain and water. After submersion in glycerin, they were then read with a stereoscopic microscope by the use of reflected and transmitted light. Osmerid scales were processed much like those of herring, although swabs were mounted for the reading of each specimen rather than a single scale, as is the case with herring. Boreal smelt otolith masses were determined by the use of a Mettler analytical balance to the nearest .0001 gr.

Table 3. Ageing methods used on forage fish, R. U. 19, 1976-1977.

Species	Otolith	Scale	Otolith Mass	Length
Pacific herring		x		x
Boreal smelt	x	x	x	x
Capelin	x	x		x
Eulachon	x	x		x

Since herring frequently lose part or all of their body scales in handling, samples were taken opportunistically from anywhere except along the lateral line, although preference was given to the anterior portion of the body above the lateral line. Scale samples taken in the field were put into small envelopes and all pertinent data was written on the envelope. Scales from these whole fresh or frozen fish were mounted directly from the fish onto a glass slide with a dilute glue solution at the Kodiak facility (Figure 4).

Fecundity analysis was done on three forage fish species in 1977. Ovaries were dissected out of 84 herring, 68 capelin, and 6 eulachon. The ovaries were taken from ripe females, but care was taken to be sure that no partially spawned out fish were sampled. When processing began, Gilson's Fluid* was

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*See Appendix B for complete recipe and instructions for use of this preservative.

**$K = \frac{W}{L^3} \times 10^5$

Figure 4: Field laboratory facilities at ADF&G cabin, Port Heiden, field season, 1977.

decanted and the ovaries washed repeatedly in running tap water, strained through a wire sieve and broken up manually (with gloved hand) until most eggs were separated. Remaining fragments of membranous matter were removed with forceps. The eggs were then placed to drain on paper toweling in separate petri dishes; all standing water was decanted until no more remained pooled in the bottom of the dish. A single wall transite oven (Model SW-11TA) was used for drying purposes. This oven would hold twelve 10 oz. petri dishes (used for herring and eulachon eggs) and 36 syracuse plates with an inside diameter of 6.5 cm (used for capelin).

The drying process took from 12 to 18 hours at 60° C depending on the size of the ovary and the cohesiveness of the egg mass. (Often times, especially with thawed frozen samples, the eggs did not separate well. These clumps of eggs took longer to dry than those which were well separated.) Frequently several samples would dry before the rest, at which time more wet samples were placed in the oven so it was kept full at all times. It was necessary to be sure each sample was evenly dried throughout so that subsample weights would be reliable indicators of the whole sample. The eggs were dry when they became hard and pebble-like in texture. After drying, the eggs were placed in small carefully labeled envelopes and sealed at the time the weighing process was begun. The eggs were counted out in plastic petri dishes (14 cm in diameter) (Figure 5). It was found that herring eggs were counted most efficiently on a light background while those from eulachon and capelin were most easily counted against a dark background; three subsamples of 250 eggs were counted out from each ovary. These samples were weighed to the nearest 0.1 mg. on the analytical balance (Figure 6); whole samples were then weighed also to the nearest 0.1 mg., with the total egg count extrapolated from the mean weight of the three 250 egg samples.

Macro-analysis of stomach contents were made on boreal smelt in the field, the contents identified to order and percent fullness recorded for that specimen. Herring, capelin and eulachon, due to their advanced pre-spawning state had, for the most part, empty foreguts.

RESULTS

Aerial Surveys

During the 1977 field season 26 survey flights were made over a total trackline of 13,262 kilometers with single and twin engine aircraft for the specific purposes of this Research Unit. All flights took place between Cape Sarichef on the western end of Unimak Island northeast along the Bering Sea coast to the mouth of the Yukon River. Over this area a total of 1,118 schools of forage fish were sighted during 52 individual surveys. Eighty-five percent of all schools were seen during the last week of May and the first week of June. Because of the extensive length of this coastline, it was broken into three sub-areas (Table 4), which can be seen on the following page. The areas of forage fish concentrations as seen during aerial surveys are represented in Figure 7, and the computations derived from these surveys are presented in Table 5. Greatest sighting rates took place between Naknek and Cape Newenham, which encompasses the intensive Togiak herring fishery. Surveys between Cape Sarichef and Naknek on the Alaska Peninsula, resulted in the lowest schools seen per kilometer flown.

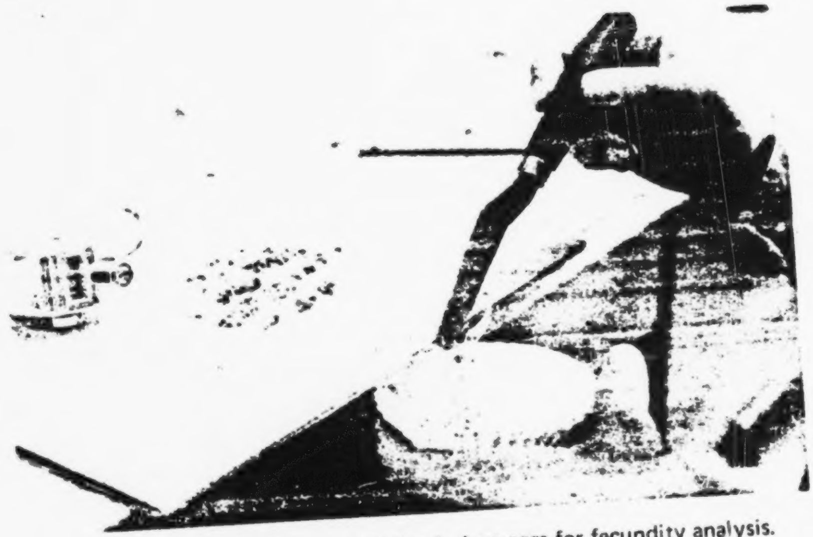

Figure 8. Counting process of dried eulachon eggs for fecundity analysis.

Figure 9. Mettler Analytical balance used for weighing dried fish eggs in fecundity processing.

Table 4. Sub-areas, forage fish aerial surveys. R. U. 19, 1977.

Area A: Cape Sarichef to Naknek

Area B: Naknek to Cape Newenham

Area C: Cape Newenham to Yukon River

Table 5. R. U. 19 Aerial census results, OCSEAP Forage Fish. Cape Sarichef to Yukon River, 1977.

(1)	Overall kilometers flown	13,262
	Overall schools sighted	1,118
	Overall schools seen/kilometers flown	.083
	Flights made	26

AREA A

(2)	Overall kilometers flown	5,403
	Overall schools sighted	59
	Overall schools seen/kilometers flown	.011
	Flights made	8

AREA B

(3)	Overall kilometers flown	3,731
	Overall schools sighted	765
	Overall schools seen/kilometers flown	.205
	Flights made	10

AREA C

(4)	Overall kilometers flown	.4128
	Overall schools sighted	294
	Overall schools seen/kilometers flown	.071
	Flights made	8

In 1976 a total of 1,481 schools of forage fish were observed over the same trackline. Thirty-four survey flights were flown over 16,565 kilometers for an overall result of .0894 schools of forage fish seen per kilometer flown, virtually identical to the .083 schools per kilometer in 1977.

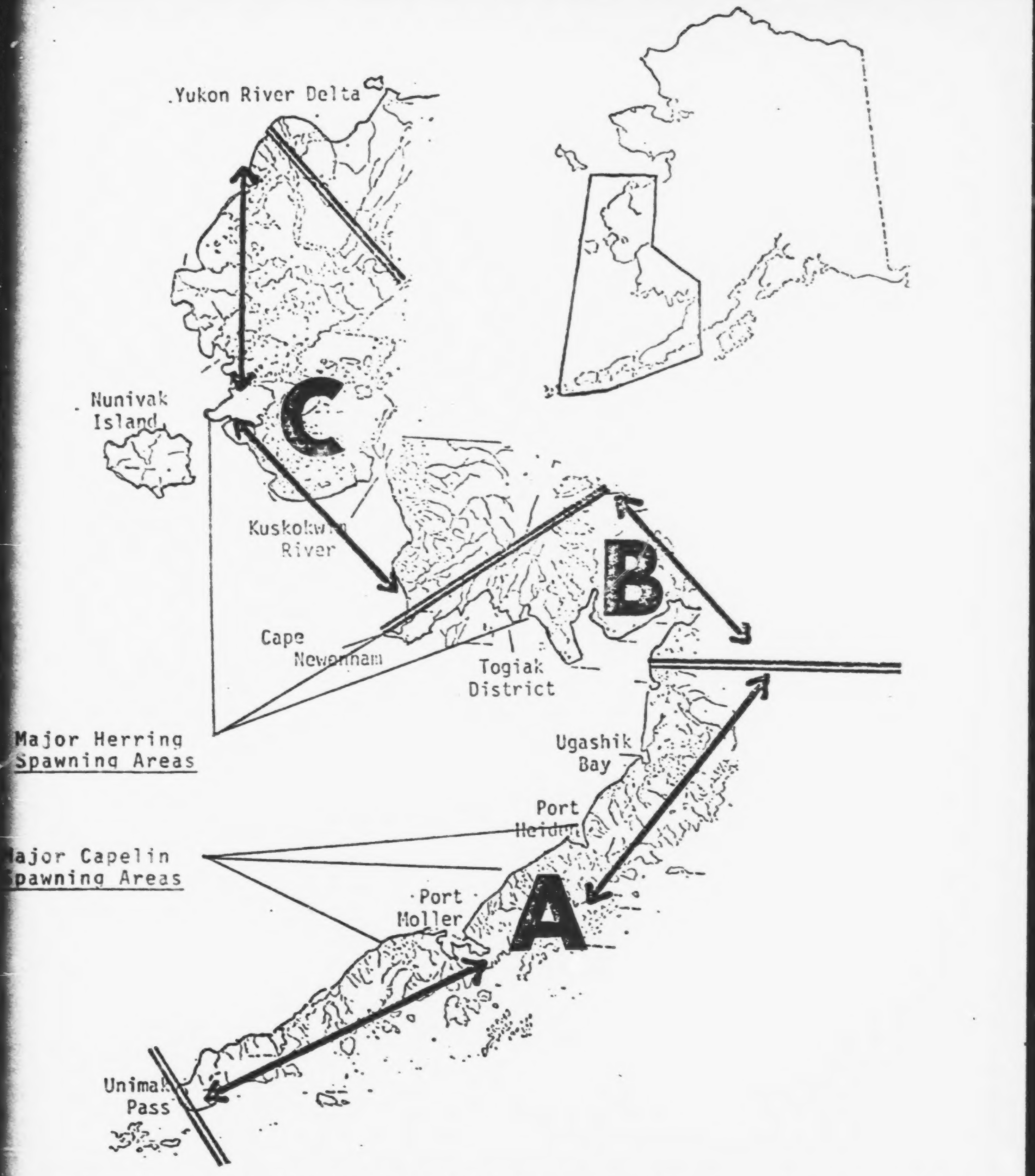
Yukon River Delta
Nunivak Island
C
Kuskokwim River
B
Cape Newenham
Togiak District
Major Herring Spawning Areas
Ugashik Bay
Port Heiden
Major Capelin Spawning Areas
Port Moller
A
Unimak Pass

Figure 7. RU 19 (Extension) 1977 Study area, showing major geographical points and major forage fish spawning areas.

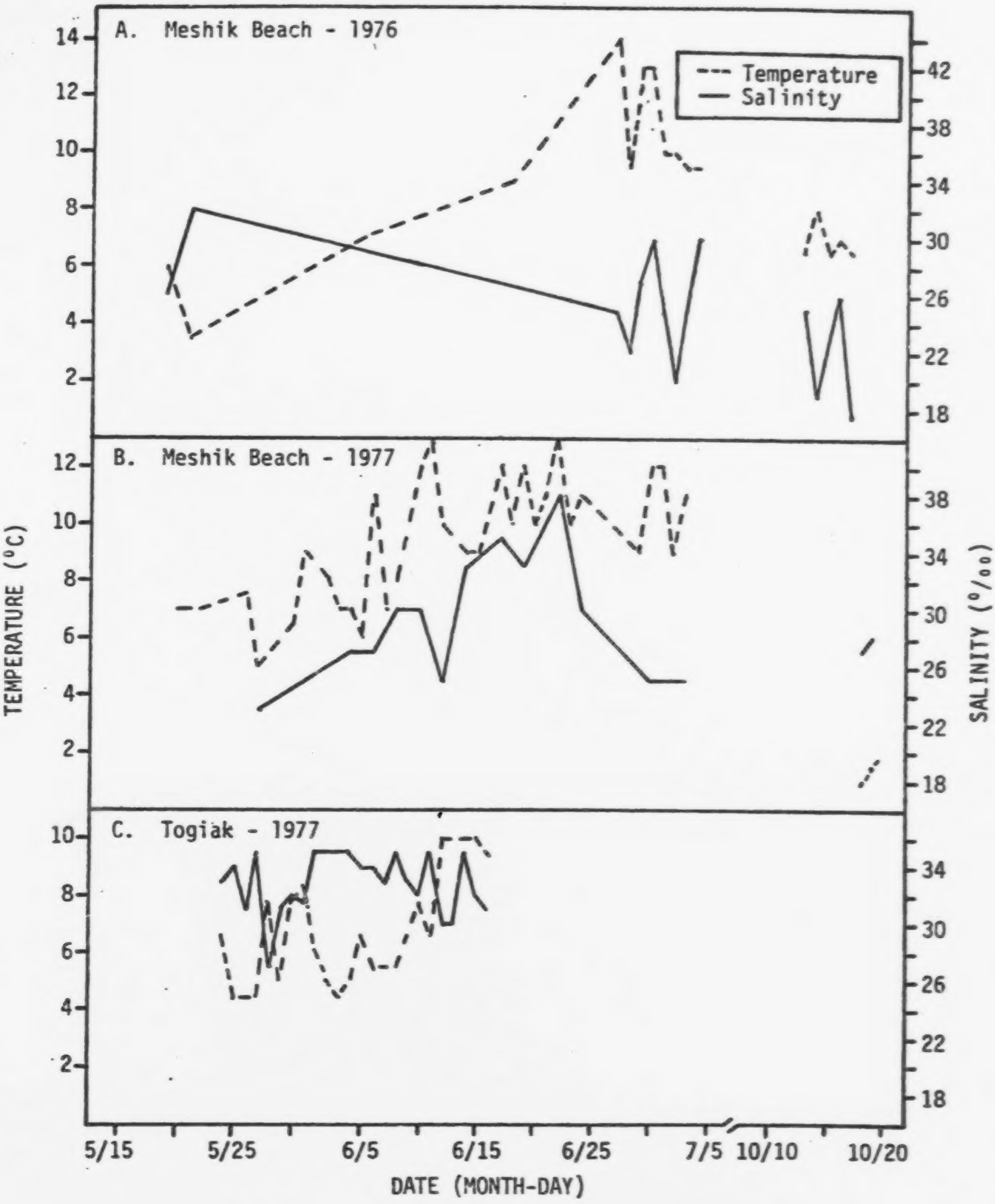

FIGURE 8. Temperature and salinity data obtained from three primary forage fish sampling locations on the north Alaska Peninsula and Bristol Bay during the 1976 and 1977 field seasons.

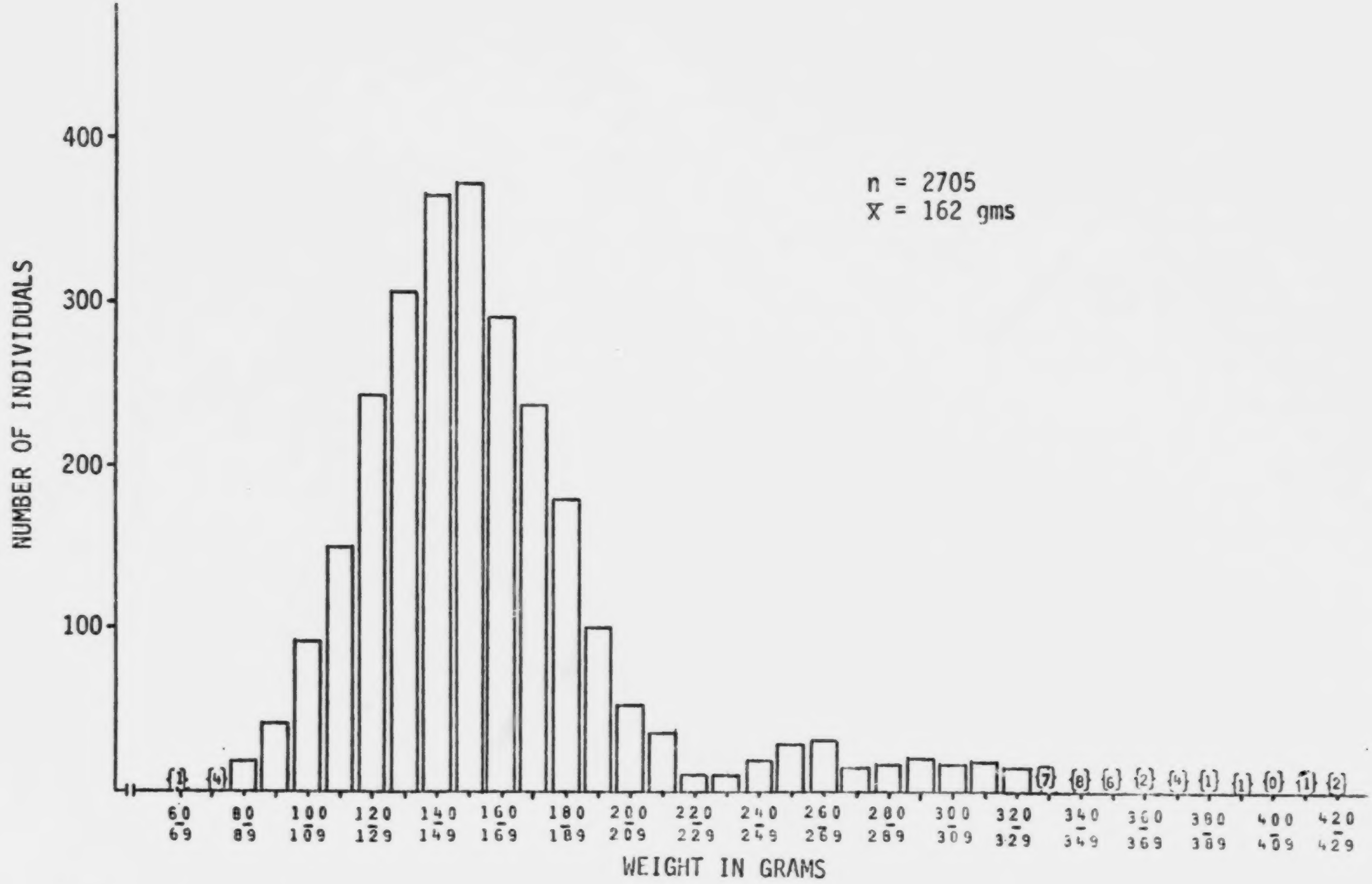

FIGURE 9. Weight frequency distribution of Pacific herring from Togiak district, 1977.

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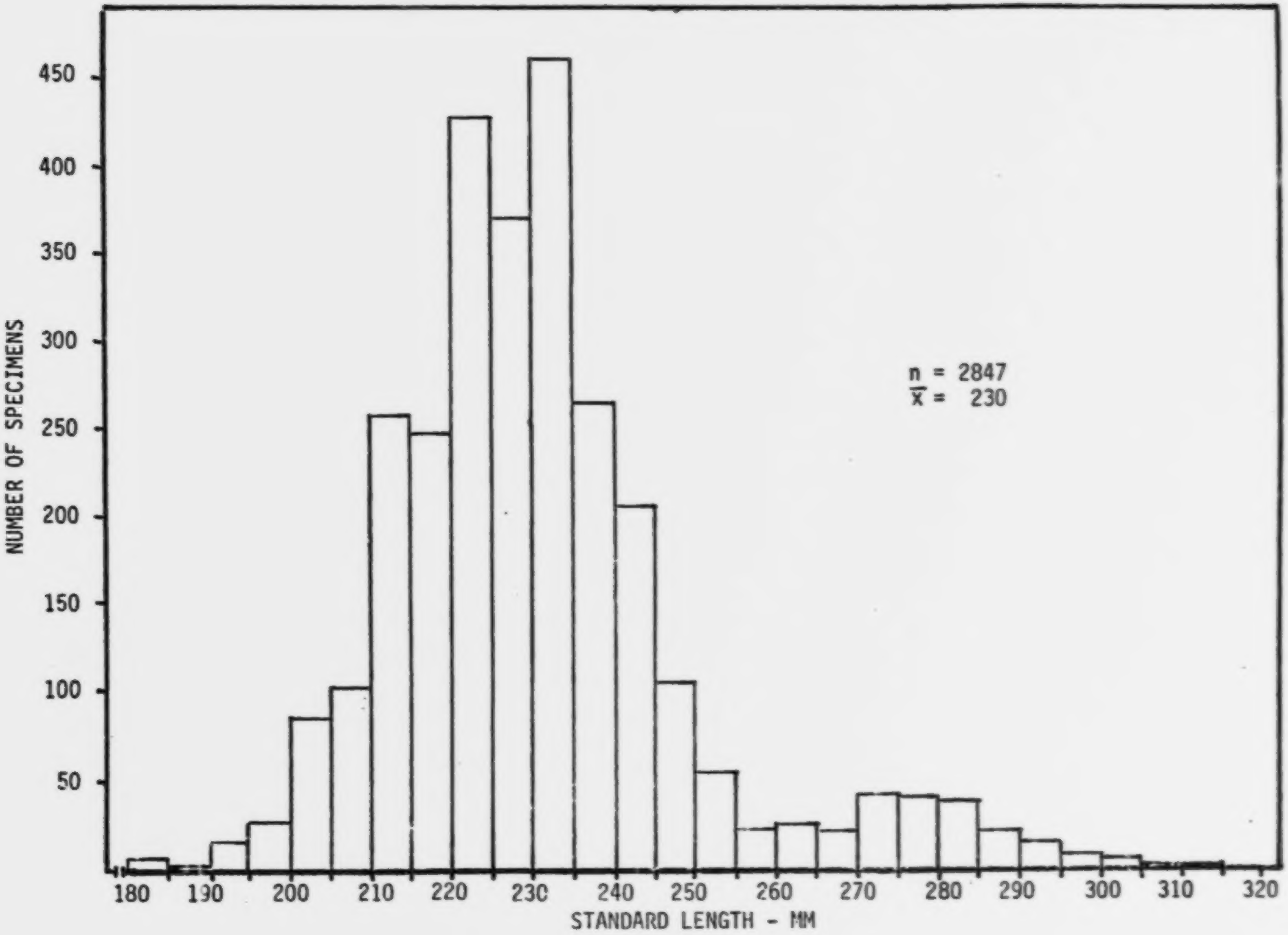

FIGURE 10. Length frequency distribution of Pacific herring from Togiak district, 1977.

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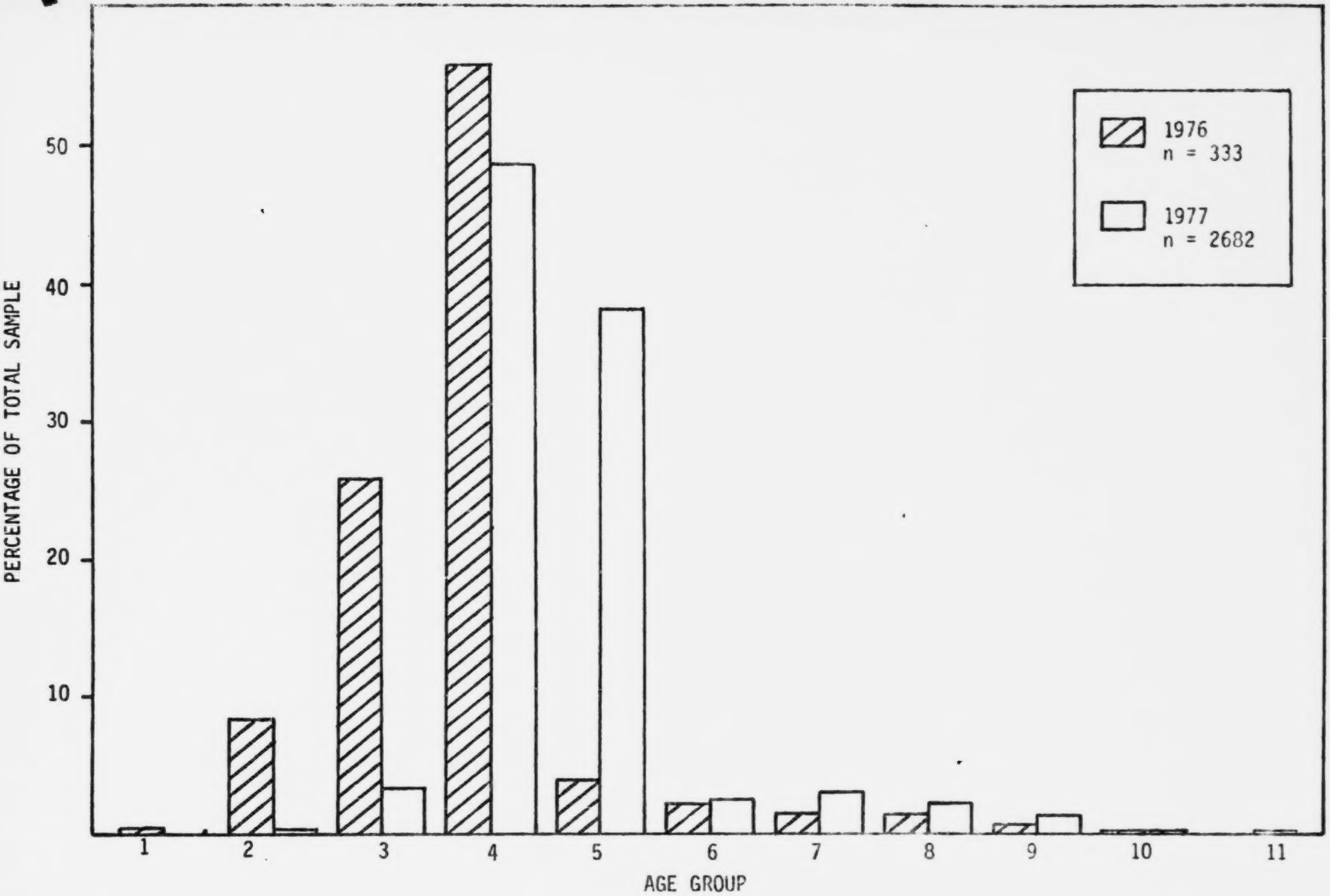

FIGURE 11. Age group composition of Pacific herring taken in the Bristol Bay area during 1976 and 1977.

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Hydrography

Thirty-four temperature readings and 31 salinity readings were taken during 1976 in the contract area. In 1977, however, salinity data was restricted to Togiak Bay and Meshik-Port Heiden. A total of 24 temperature and salinity readings were taken in Togiak Bay, while 37 temperature and 17 salinity readings were taken at Meshik-Port Heiden. Temperature increased from about 4° to 8° C. in late May, to 9° to 14° in late June at Meshik each year, but was cooler at Togiak. Salinity fluctuated from about half to full seawater strength at Meshik but was generally full seawater salinity at Togiak (Figure 8).

Fish Sampling

Summary

A total of 4,738 forage fish were taken for the purposes of the RU in 1977. The majority of these were herring (2,682) most of which were taken at Togiak Bay in May from commercial purse seine catches.

A total of 1,416 capelin were taken, 108 from Meshik-Port Heiden, and 1,308 from Togiak Bay. All boreal smelt captured (405) in 1977 were taken at Meshik-Port Heiden, as were the 35 eulachon.

Herring - Results

Herring were first captured on May 7 at Togiak and on May 23 at Meshik-Port Heiden. The mean body weight and length of sampled herring was 162 grams and 230 mm respectively. Male and female herring were equally abundant and did not differ in size. The weight frequency yielded a range of 80 to 329 grams (Figure 9). The length frequency yielded a range of 180 to 305 mm (Figure 10).

The mean age of 2,674 herring was 4.7 years, with the fourth year class predominating. Eight year classes were found. Age composition of 3,015 herring in the Bristol Bay area during 1976-77 indicated age 3 and 4 herring were abundant in 1976, and age 4 and 5 were prominent in 1977 (Figure 11). Male and female herring were equally abundant and did not differ in size.

The mean fecundity of 84 herring from Togiak was 25,350 ova with a standard error 2,377 (Table 6). Since the fecundity samples were handled in three somewhat different ways, each group was regressed separately against fecundity so as to show possible individual variances (Figure 12). Fecundity from 1976 and 1977 agree closely as mean fecundity figures between the two years are within 2 percent (Tables 6 and 7).

Inspection of herring sexual maturity indicated that most were 4 and 5 years old at maturity and some matured during their third year, but none during their second.

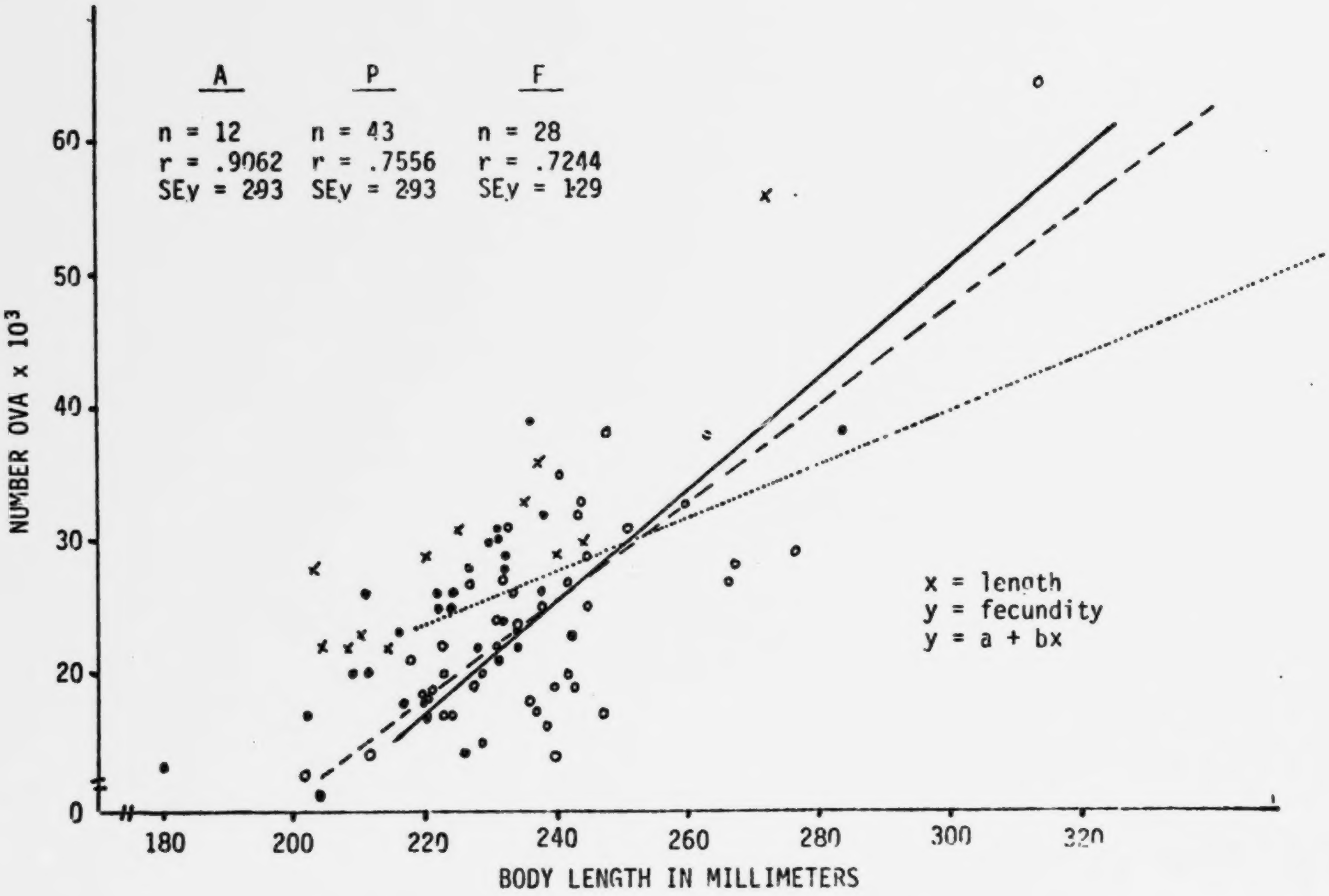

FIGURE 12. Three length/fecundity regression groups, Togiak herring, 1977.

Table 6. Mean herring fecundities in thousands of eggs by size; Togiak 1977.

Standard Length Millimeters	Mean Fecundity	Standard Deviation	Number of Fish
171-180	13.1	--	1
181-190	--	--	0
191-200	--	--	0
201-210	19.6	6.80	8
211-220	20.9	4.11	10
221-230	23.7	5.33	20
231-240	25.9	5.91	26
241-250	26.8	6.67	11
251-260	27.8	--	1
261-270	31.2	5.99	3
271-280	42.4	15.63	2
281-290	38.5	--	1
291-300	--	--	0
301-310	--	--	0
311-320	63.6	--	1
Mean	25.35	5.69	84

Table 7. Mean herring fecundities in thousands of eggs by size from Bristol Bay - Bering Sea in 1976.

Standard Length Millimeters	Mean Fecundity	Standard Deviation	Number of Fish
171-180	--	--	0
181-190	--	--	0
191-200	19.4	4.60	13
201-210	20.7	7.03	8
211-220	21.3	5.98	12
221-230	27.8	4.03	8
231-240	28.9	4.88	7
241-250	32.9	2.12	2
251-260	40.4	--	1
261-270	45.9	8.64	3
271-280	71.9	19.0	2
281-290	--	--	0
291-300	--	--	0
301-310	--	--	0
311-320	--	--	0
321-330	--	--	0
Mean	26.53	11.14	56

Linear regressions of gonad weights to body length showed a positive relationship (Figure 13).

Condition factors were determined for spring and late/summer/fall herring on 60 specimens from the Kodiak and Togiak areas. It was determined that the condition factor is much higher for summer/fall herring than for spring herring (Figure 14). It is felt that the difference is due to the season and not to the different areas of capture.

Capelin

Port Heiden/Meshik

Spawning capelin first appeared at this site on May 30, 1977 and left on June 14, 1977. Mean body length and weight of the 108 capelin was 144 mm and 21 gms respectively (Figures 15 and 16). Age of Meshik/Port Heiden capelin was 2.1 years (Figure 17). Female and male mean length were identical.

Togiak Bay

The 1,308 capelin captured averaged 136 mm long and 18.5 gms in weight. Age results from 105 otoliths yielded a mean age of 2.1 years (Figure 18). Results show the males were larger than females (Figure 19). Fecundity of 68 Togiak capelin ranged from 6,200 to 19,900 ova with an arithmetic mean of 10,600 ova (Figure 20).

Boreal Smelt

Boreal smelt were caught on the first day of sampling in 1977 (May 21) and catches continued until the last day of sampling (July 4). Boreal smelt averaged 155 mm in length and 21 gms in weight (Figures 21 and 22). Scales of 175 specimens were examined resulting in a mean age of three years (Figure 23). Mean lengths of females and males showed no appreciable difference, the former being 154 mm and the latter 155 mm. A total of 260 smelt foreguts were examined, and revealed that mysids make up the predominant portion of the animal's diet during spring (Figure 24).

Scale topography of two different age classes was studied on 238 boreal smelt. Three-year old boreal smelt showed two groupings in the number of circuli before the initial annuli. Two-year old smelt did not form two groups, rather a single one (Figure 25).

Eulachon

Small catches of eulachon between June 5 and June 29 at Meshik-Port Heiden resulted in a mean length of 224 mm and mean weight 94 gms (Figure 26). Age determinations from otoliths showed a mean age of 3.1 years (Figure 27). Mean male-female body length differences were slight, with males being 223 mm in body length and females 227 mm. Fecundity samples from six specimens yielded a mean fecundity of 41.9 thousand ova, the range being from 34,000 to 57,000.

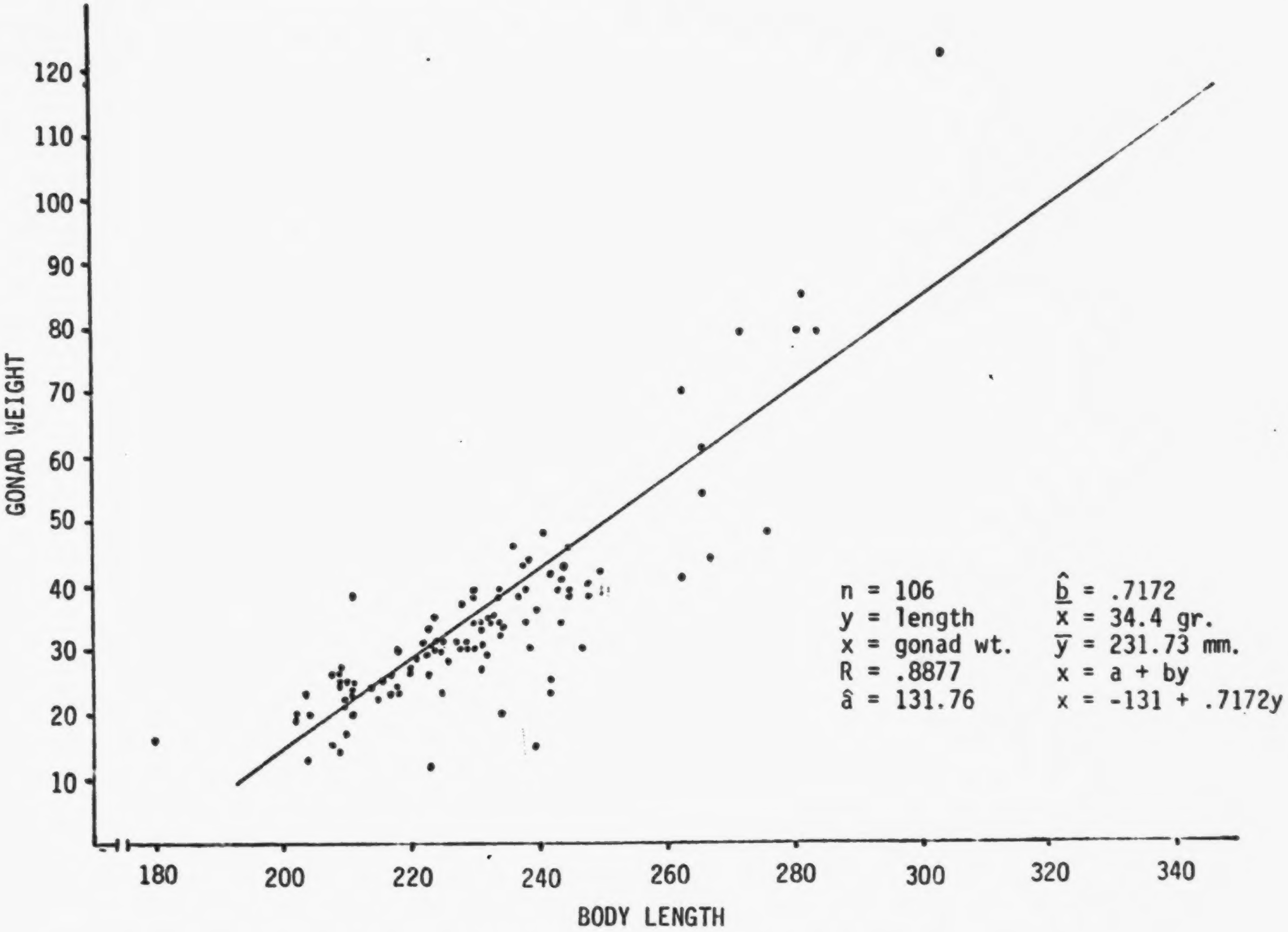

FIGURE 13. Herring body length plotted against gonad weight; Togiak, 1977.

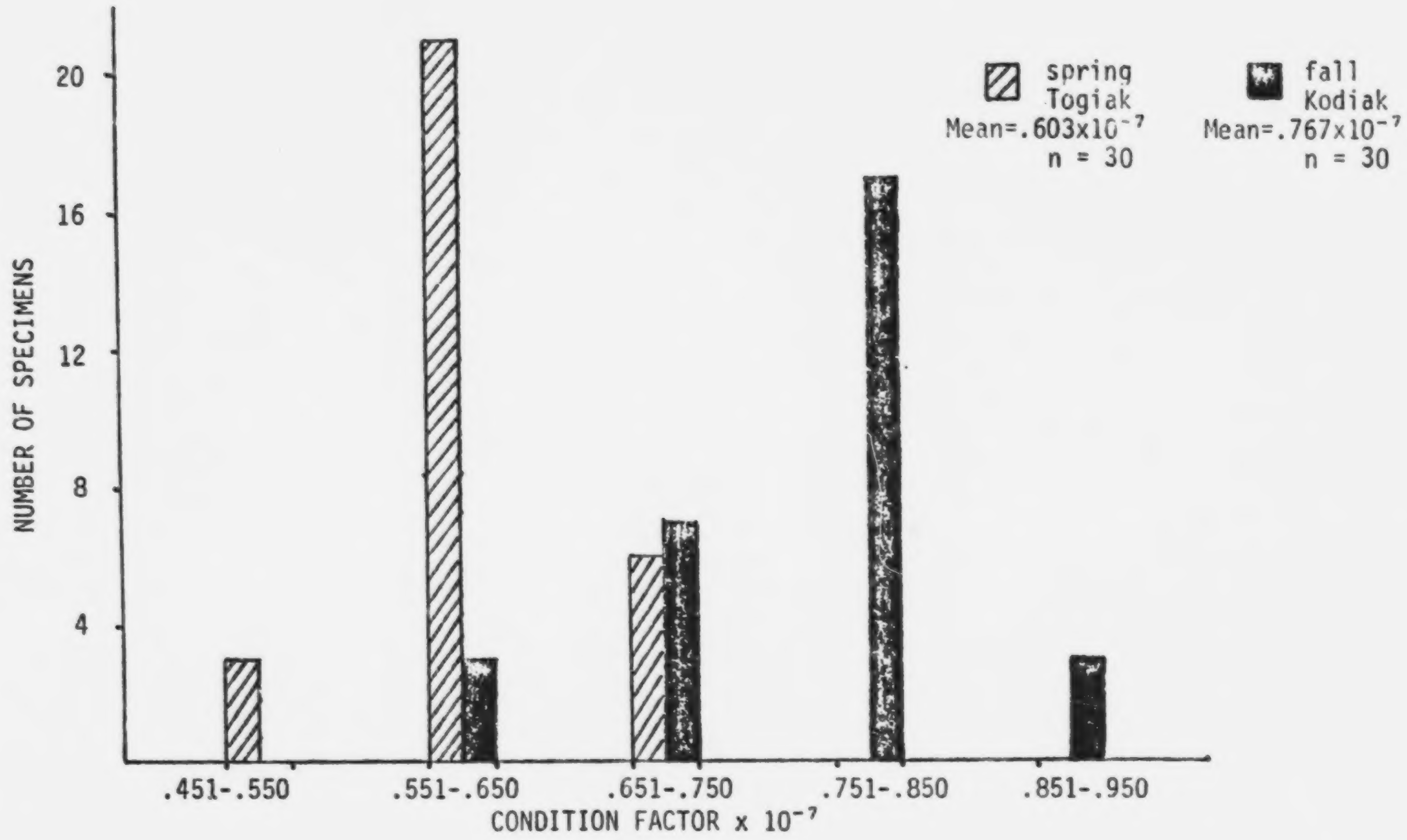

FIGURE 14. Comparative condition factors of spring and fall Pacific herring taken from Kodiak and Togiak, 1977.

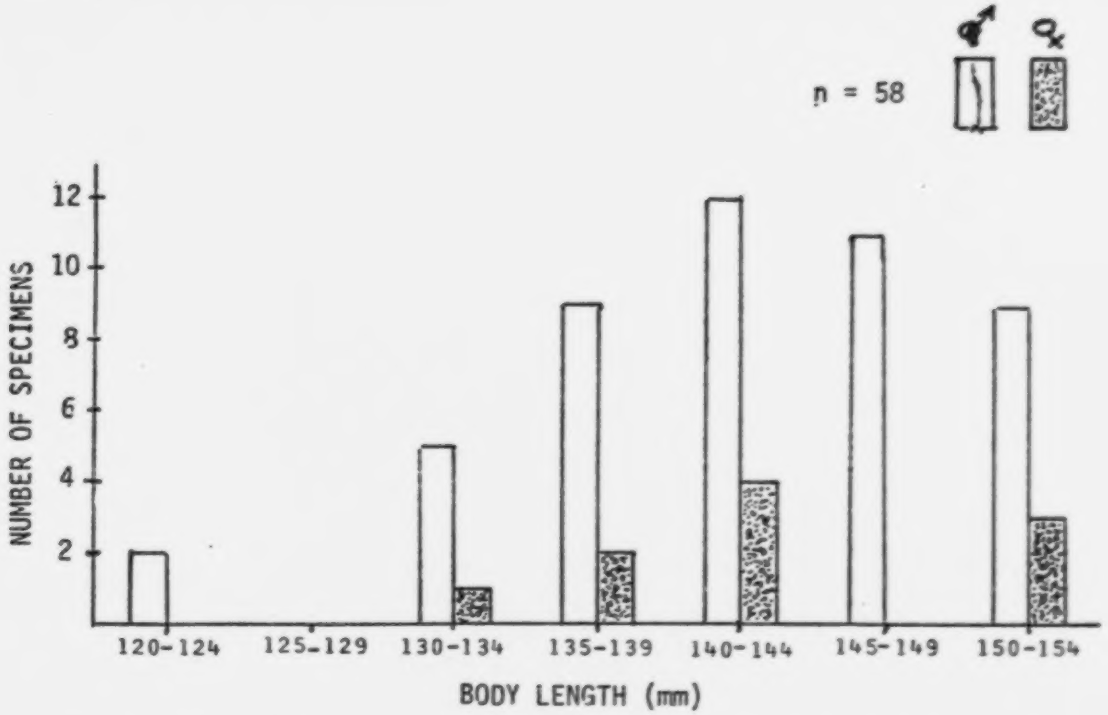

FIGURE 15. Capelin length distribution, Meshik-Port Heiden, 1977.

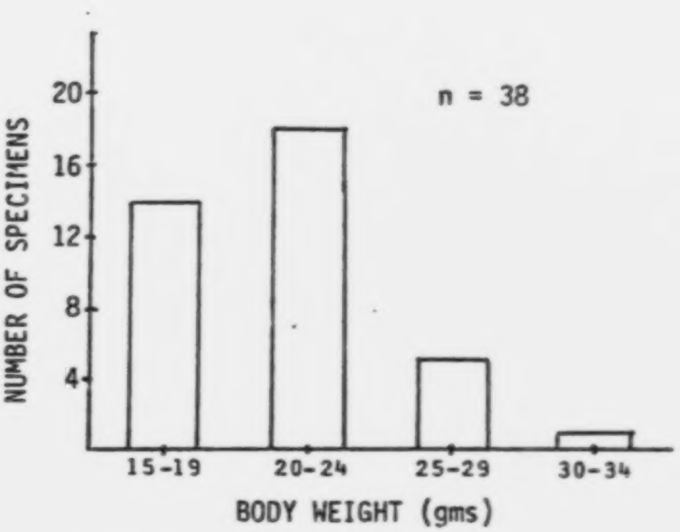

FIGURE 16. Capelin weight distribution, Meshik-Port Heiden, 1977.

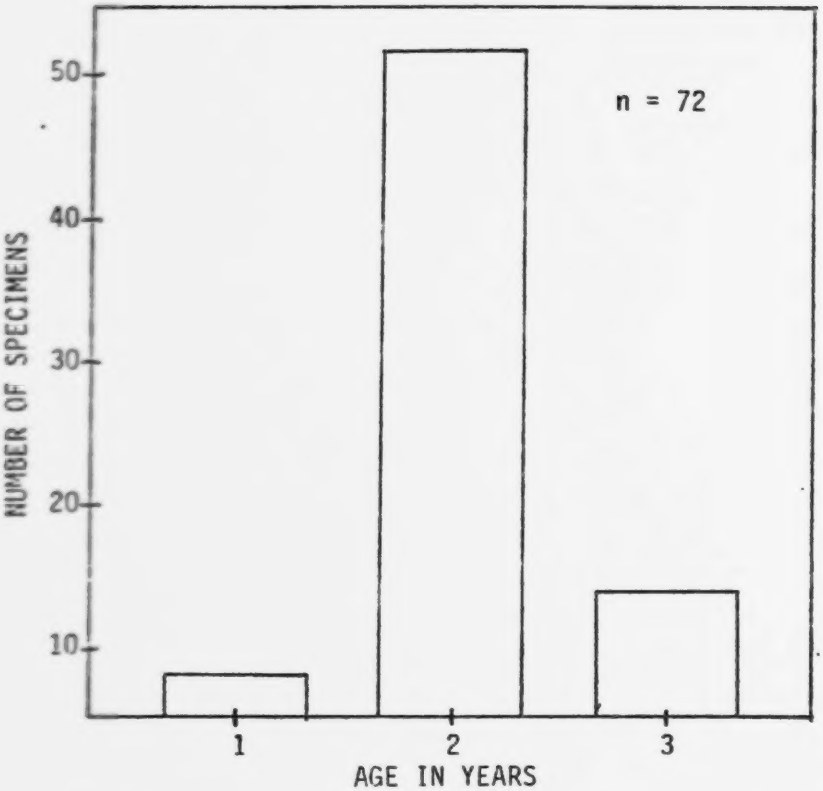

FIGURE 17. Caplin age distribution as determined from otoliths, Meshik-Port Heiden, 1977.

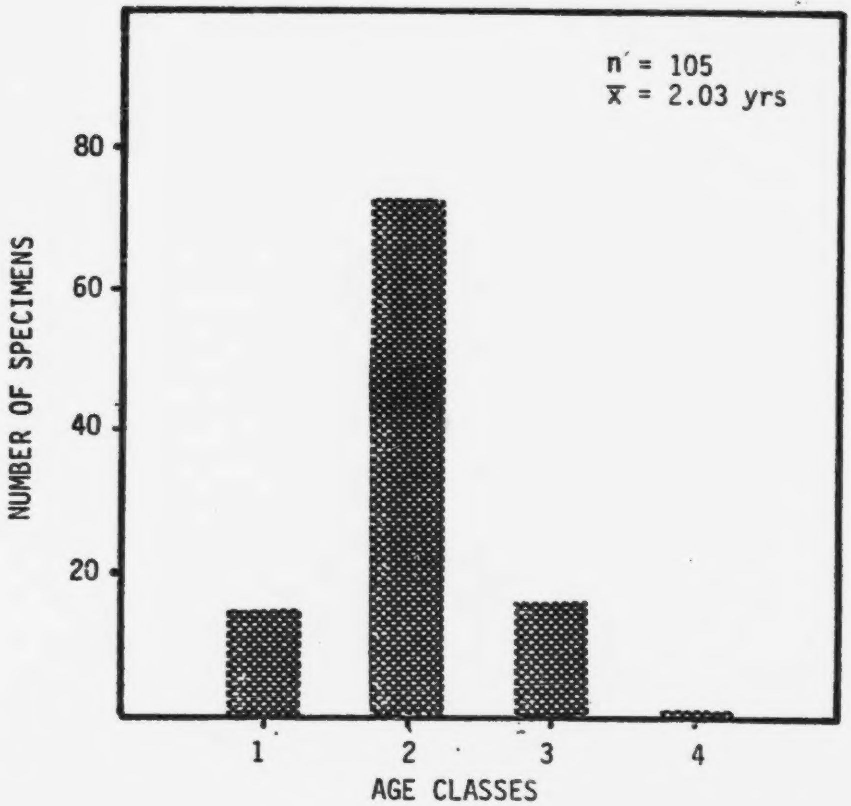

FIGURE 18. Age composition of capelin caught at Nunavarchak Bay, Togiak district, June 1977.

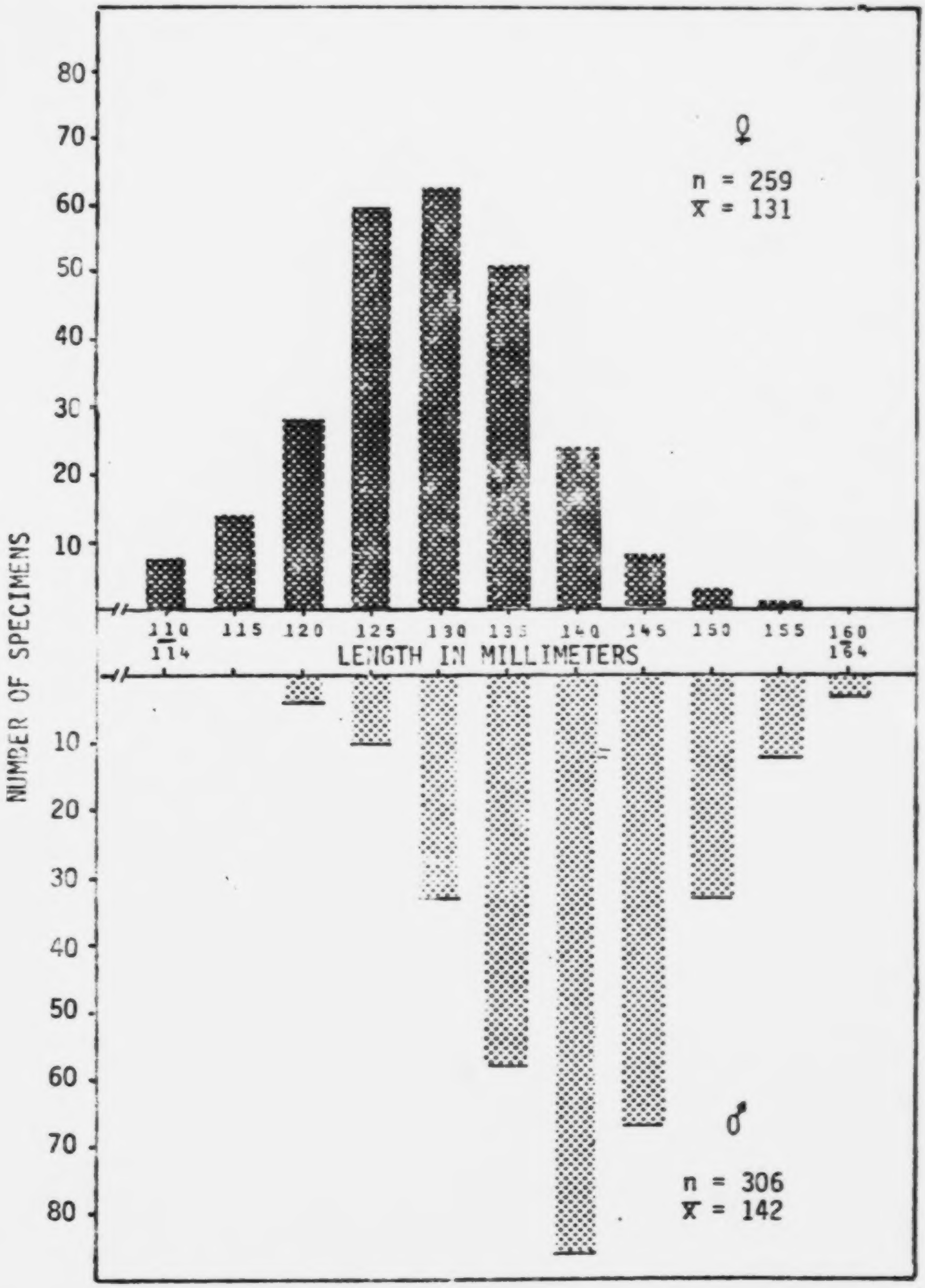

FIGURE 19. Length distribution of male-female capelin caught at Nunavarchak Bay, Togiak district, June 1977.

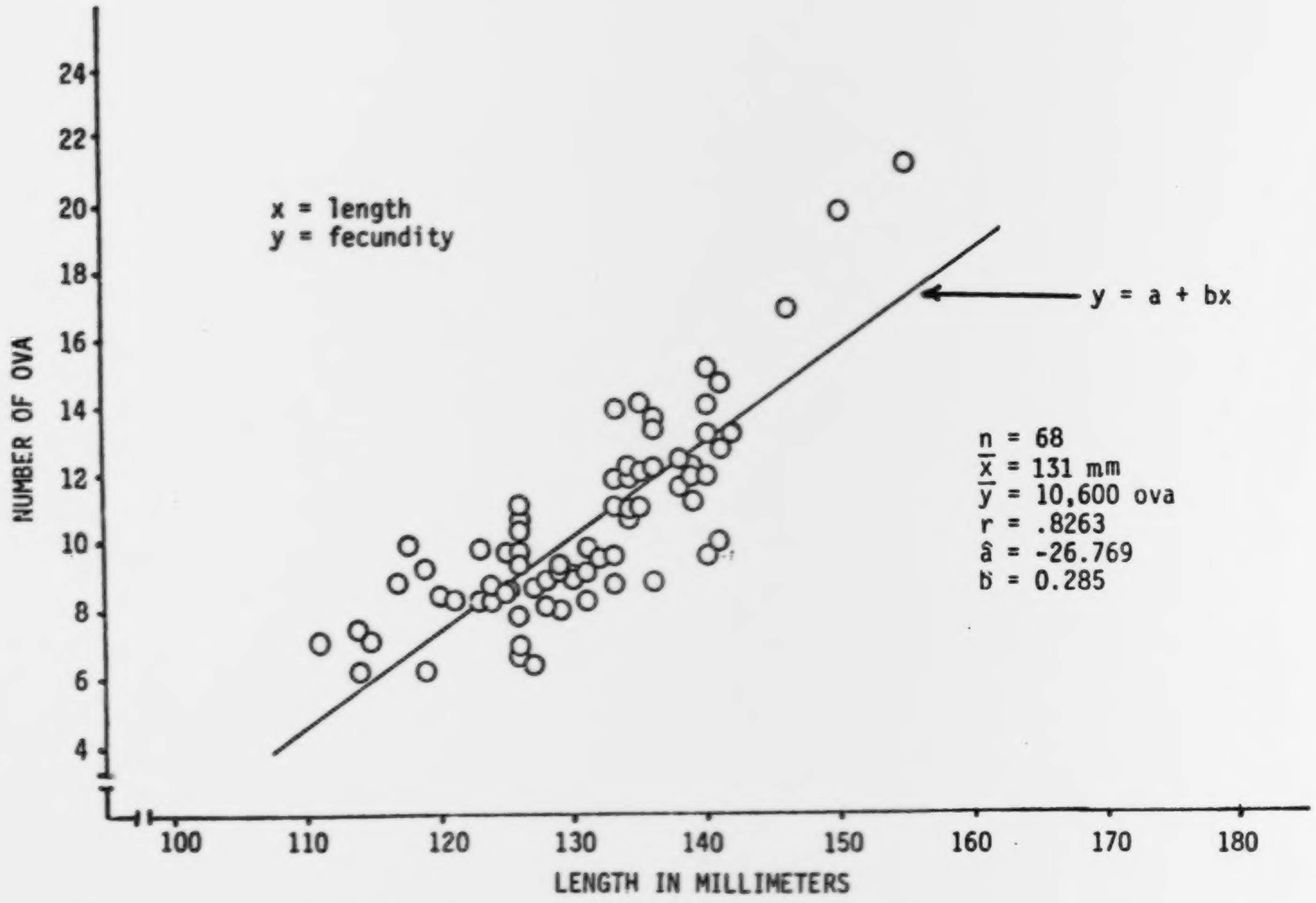

FIGURE 20. Relationship of length to fecundity of capelin from specimens taken at Nunavarchak Bay, Togiak district, June 1977.

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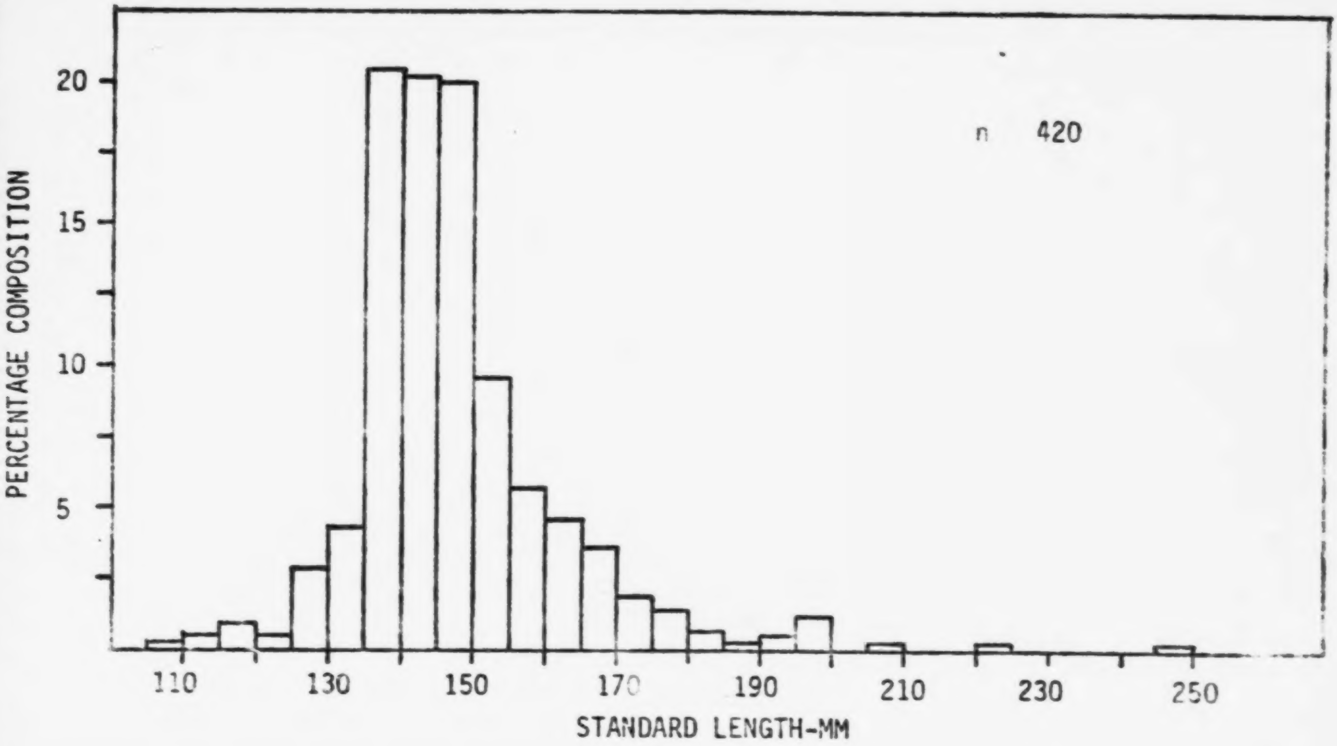

FIGURE 21. Size frequency composition of boreal smelt caught at Meshik-Port Heiden during the spring - summer 1977 field season.

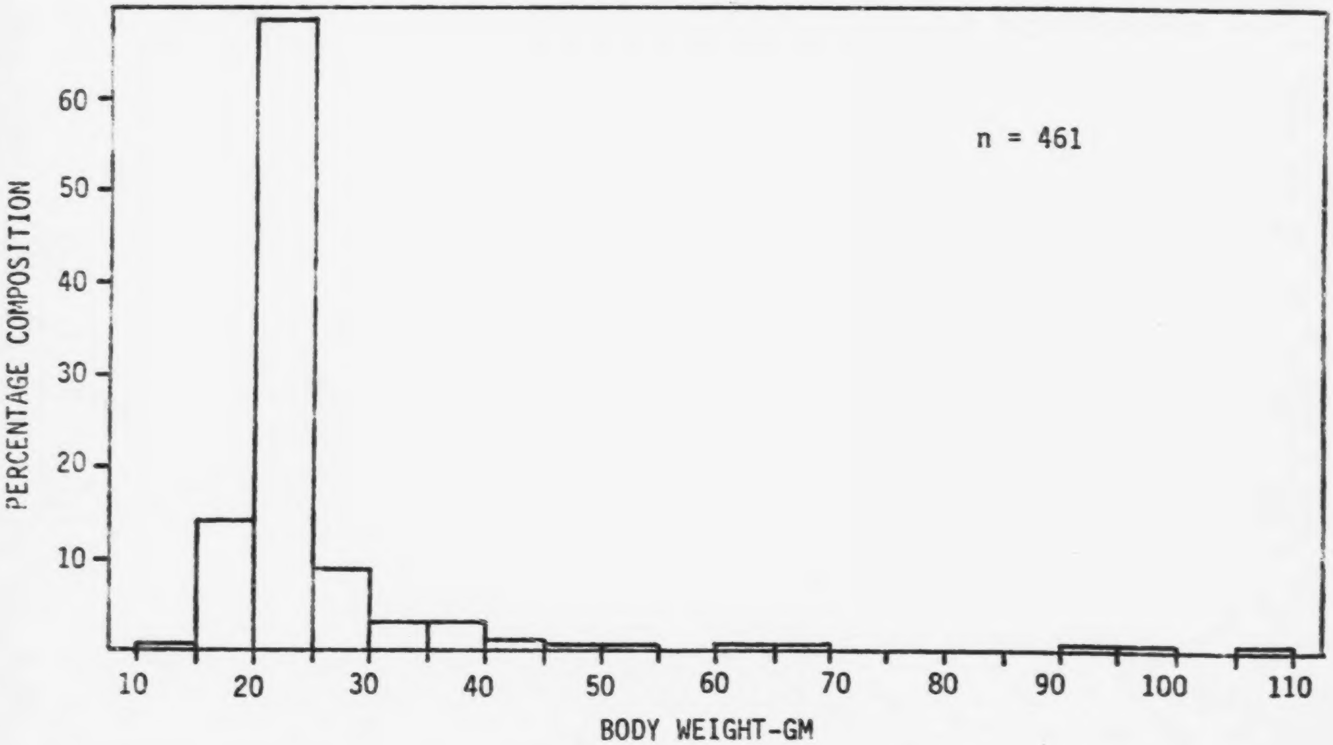

FIGURE 22. Weight frequency of boreal smelt caught at Meshik-Port Heiden during the spring - summer 1977 field season.

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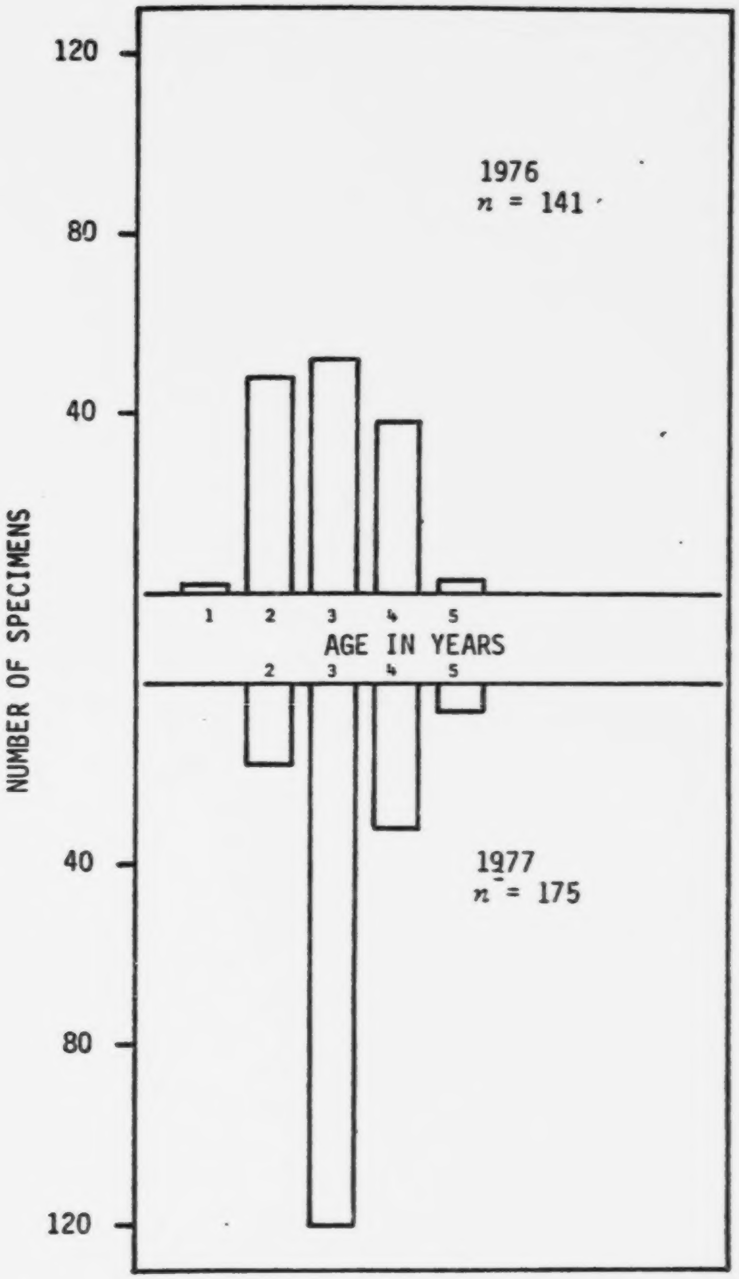

FIGURE 23. Age composition of boreal smelt caught at Meshik-Port Heiden during the 1976 and 1977 field seasons.

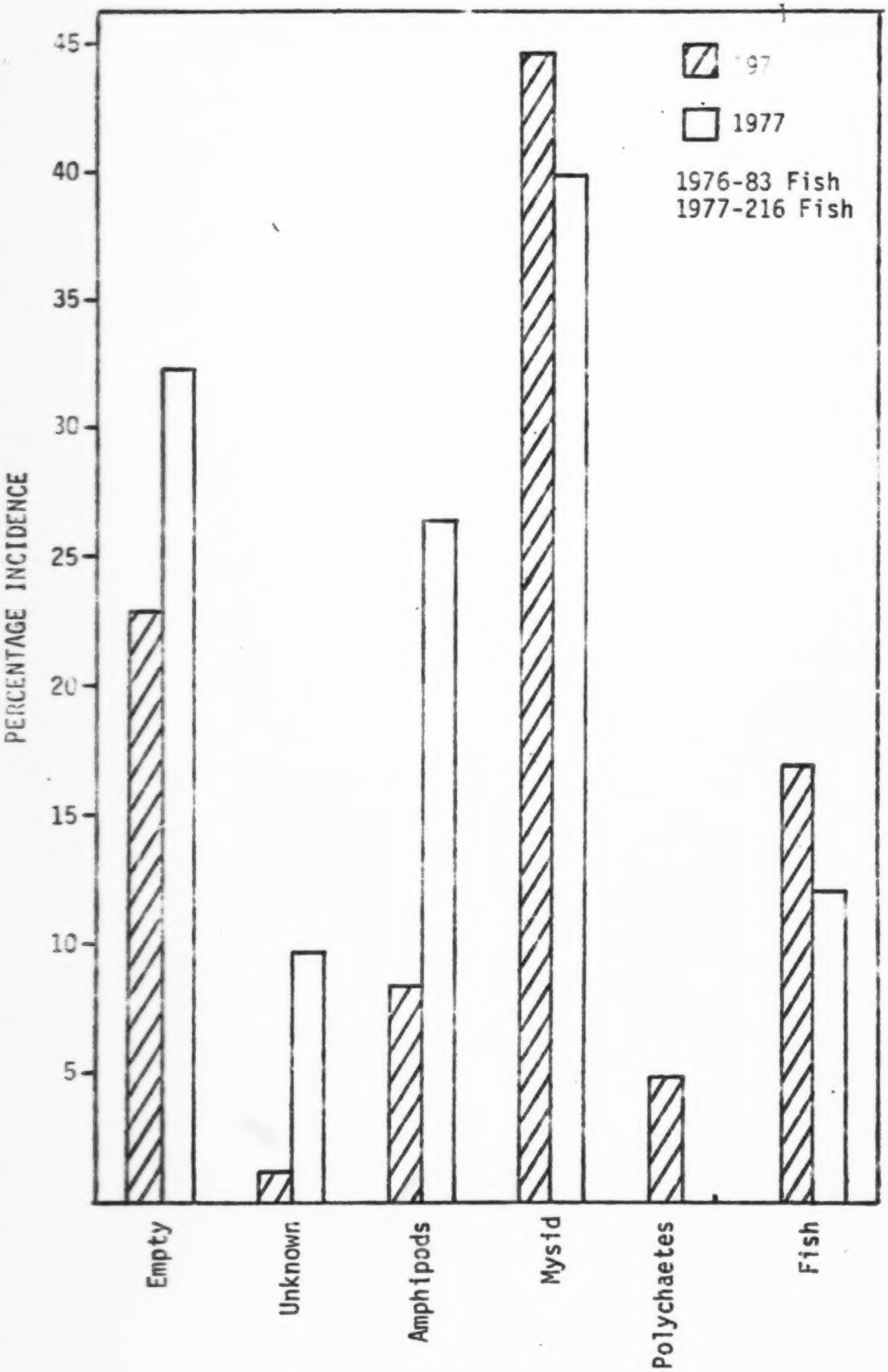

FIGURE 24. Incidence of various food items found in boreal smelt in the spring-summer of 1976 and 1977 at Meshik-Port Heiden as determined by macro-analysis of foregut contents.

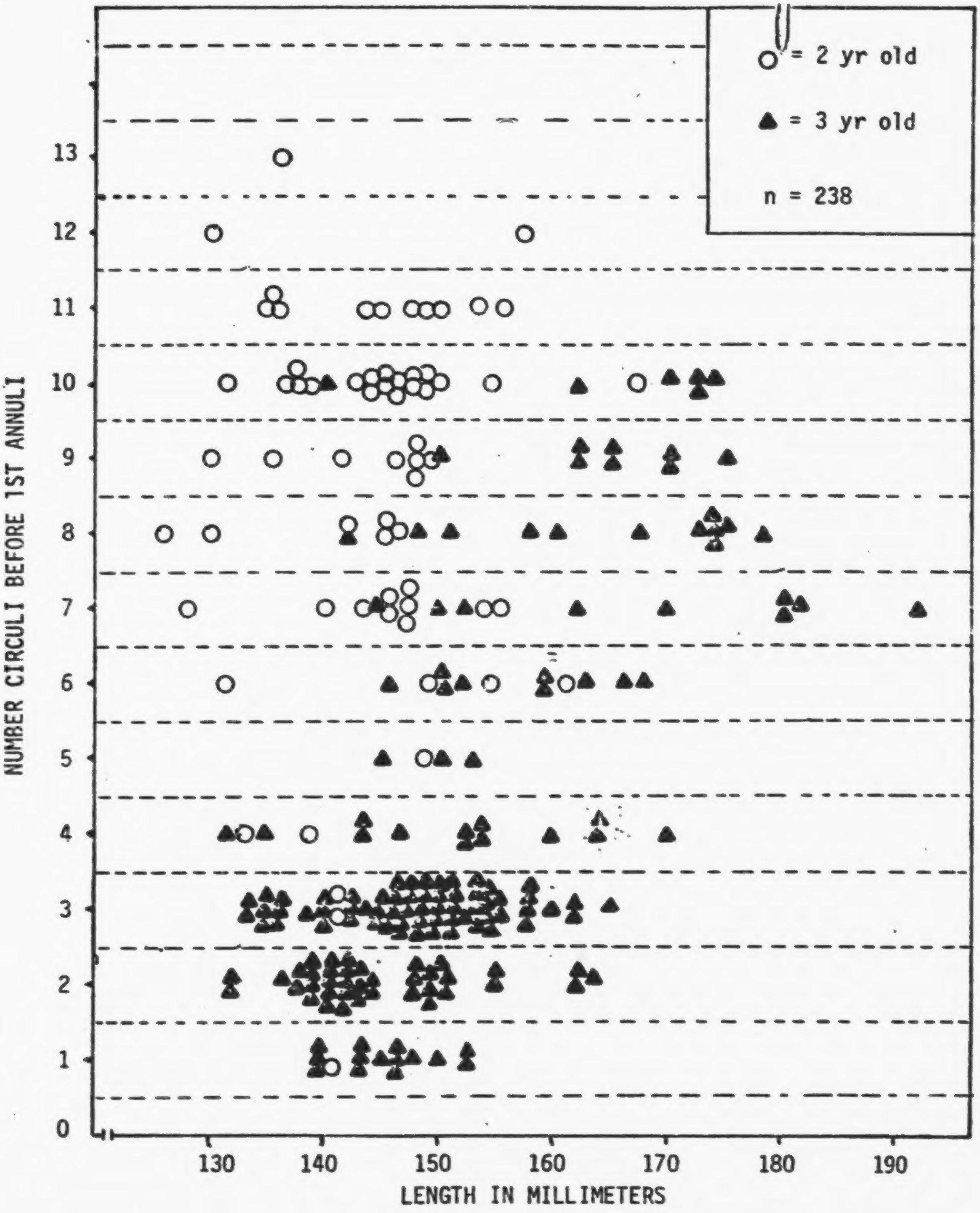

FIGURE 25. Chart showing relationship of age and size of boreal smelt to number of circuli laid down before 1st annuli in specimens caught at Meshik-Port Heiden, 1976-77.

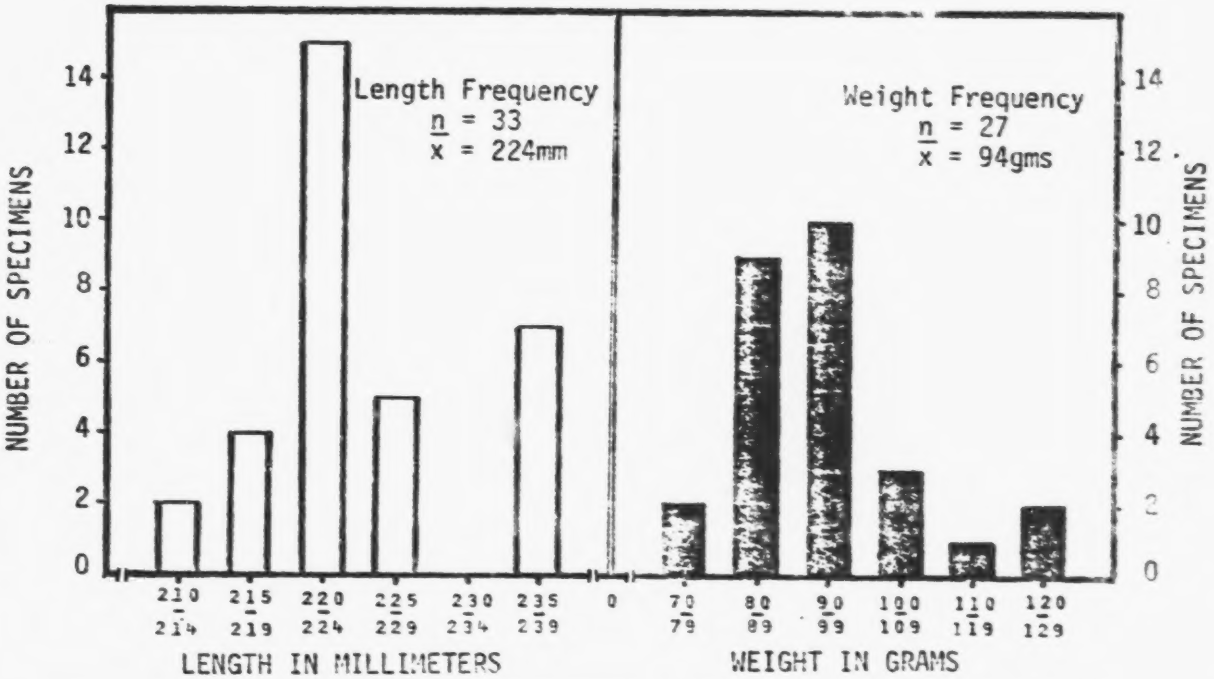

FIGURE 26. Length, weight distribution of eulachon caught at Meshik-Port Heiden, May-June, 1977.

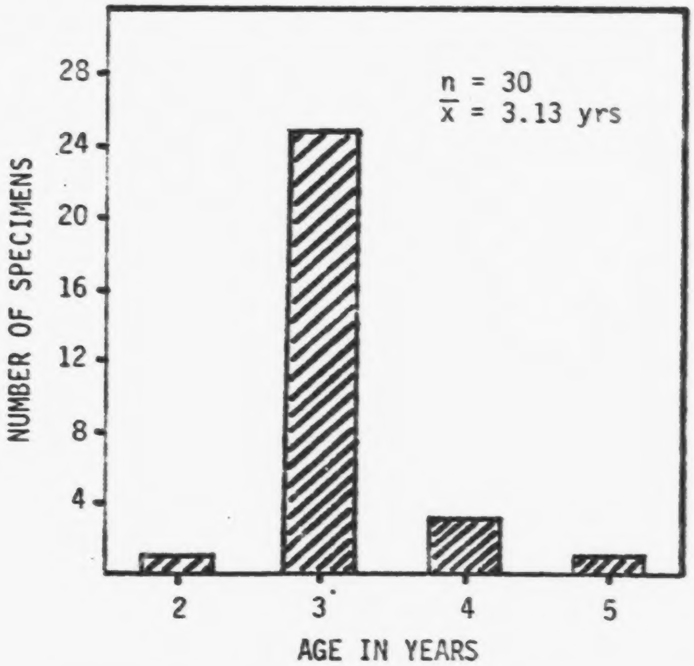

FIGURE 27. Age composition eulachon caught at Meshik-Port Heiden, May-June, 1977.

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Miscellaneous Species

A total of eleven miscellaneous species were captured incidently at the Meshik-Port Heiden test site in 1977 as opposed to 15 at this site in 1976 (Tables 8 and 9).

Table 8. Miscellaneous species caught at Meshik-Port Heiden, 1976.

Dates	Scientific Name	Area	Mean Length mm	Number Sampled	Number Caught	Data Taken
6/13-7/10	Poroclinus rothrocki	Meshik	-	-	115	-
5/16-7/10	Salvelinus sp.	Meshik	231 fl	27	60	19 otolith 9 stomachs
6/6-6/19	Salvelinus sp.	Herendeen Bay	392 fl	5	12	-
6/23-6/24	Salvelinus sp.	Seal Island	377 fl	81	85	-
6/5-7/4	Oncorhynchus kisutch	Meshik	112 fl	26	-	2 otoliths 4 scales
6/5-7/3	Oncorhynchus tshawytscha	Meshik	86 fl	2	-	-
5/21	Limanda aspera	Meshik	192 fl	1	-	-
5/23	Psettichthys melanostictus	Meshik	164 fl	1	-	otolith
5/21-7/4	Unknown	Herendeen Bay	-	-	12	-
10/31-10/20	Isposetta isolepis	Meshik	68 fl	12	-	2 otoliths
10/13-10/20	Poroclinus rothrocki	Meshik	216 fl	7	-	-
10/13-10/20	Myoxocephalus polyacanthocephalus	Meshik	191 fl	5	-	-
10/13-10/20	Hypomesus pretiosus	Meshik	124 fl	3	-	-
10/13-10/20	Ammodytes hexapterus	Meshik	162 fl	2	-	2 otoliths
10/13-10/20	Eleginus gracilis	Meshik	267 fl	1	-	otoliths
10/13-10/20	Limanda aspera	Meshik	166 fl	1	-	otoliths
10/13-10/20	Hemitripterus bolini	Meshik	235 fl	1	-	-
10/13-10/20	Cottidae sp.	Meshik	268 fl	1	-	-

Table 9. Miscellaneous species caught at Meshik-Port Heiden, 1977.

Dates	Scientific Name	Area	Mean fK Length mm	Number Caught	Number Sampled	Data Taken
5/21-6/22	*Liopsetta glacialis*	Meshik	180	21	-	-
	Platichthys stellatus	Meshik	215	16	-	-
	Poroclinus rothrocki	Meshik	238	6	-	-
	Pungitius pungitius	Meshik	-	5	5	-
	Enophrys claviger	Meshik	187	3	-	-
	Dasycottus setiger	Meshik	174	3	-	-
	Salvelinus sp.	Meshik	118	2	-	-
	Oncorhynchus tshawytscha	Meshik	119	2	-	-
	Ammodytes hexapterus	Meshik	156	1	1	otolith
	Hexagrammos sp.	Meshik	112	1	-	-
	Eleginus gracilis	Meshik	178	1	-	-
	Unknown	Meshik	-	-	21	-

DISCUSSION

Herring

General Life History

Herring are a completely marine species of clupeid which mature at age three or four in the Bering Sea, and frequently live to be eight or more years of age. Atlantic and Pacific herring have the same outward appearance except for the position of the scutes* on the underside of the fish (i.e. in the Pacific herring the scutes do not extend anterior of the ventral fins as they do in Atlantic herring). Pacific herring were thought to spawn only along vegetated nearshore zones in the spring, whereas Atlantic herring spawn either in spring, fall or winter in deep water, offshore areas (Jones, 1968).

The distribution of the Pacific herring extends easterly in the Arctic Ocean to Cape Bathurst on the coast of the Northwest Territory and westerly to the Kara Sea in northern Russia; the southern limit of their range appears to be San Diego Bay on the North American coast and as far south as the East China Sea on the Asian continent.

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*Scutes: A bony, chitinous external plate or scale, in this case scales on the underside of the fish which form a dorsal line or protrusion.

Discussion

Rumyantsev and Darda (1970) studied fecundity of herring in the Bering Sea; our 1976-77 results are comparable to those found by these Russian investigators. Multiple regressions were calculated on both 1976 and 1977 herring fecundity samples, with variables being length, weight and age. The results from this analysis were inconclusive. In previous fecundity research, body length normally gives the highest correlation in predicting relative fecundity (Nagasaki, 1958; Messieh, 1976; Pitt, 1965; etc.). Since our results indicated that gonad weight was related to increased body length, one would anticipate fedundity increasing with the latter parameter, but that proved not to be necessarily the case. Often times the fish with larger gonad size prove to be less fecund.

The point in question, is that 1976 fecundity results completed by OCS investigators in the Bering Sea agreed precisely with the previous fecundity work completed on herring (Rumyantsev & Darda, 1970); also, increased body weight correlated well with increased fecundity. In contrast to this, 1977 fecundity results showed a degree of variability as egg samples were preserved in different ways, as opposed to 1976 when all ovaries were fixed identically. In 1977 collected gonads were frozen whole, then processed later; immediately fixed in Gilson's Solution, or immediately fixed with methyl alcohol. It is suggested that if similar studies are undertaken again, that identical methods of preservation be used on all herring gonads collected.

Since the condition factor of spring herring is lower than late summer or early fall herring, an oil spill in the fall would find the animal in a robust condition with much stored body fat, i.e. able to go long periods without food. In contrast, late winter, spring or early summer impact would catch the animal at a low point in respect to body condition. Hence, a spring oil spill presents the greatest risk because herring are in the nearshore areas spawning, are in a weakened body state and probably would be unable to migrate long distances (to avoid an area of impact) without food.

Pacific herring are generally considered to return to the same spawning location each year (Jones, 1968). Our aerial surveys in 1977 showed a failure of the herring "run" in the Port Moller-Herendeen Bay area. In 1976, numerous schools of herring spawned in that complex. This complete "no show" of herring in this system could be due to one of the following reasons: 1) total fishing and natural mortality of all age classes, 2) the herring from this complex skipped spawning, staying at sea to feed, 3) the herring from this complex migrated to another area to spawn, 4) these herring spawned in deep water, and 5) the herring spawned in April immediately after "ice out".

Aerial observations were intense in this area between the latter part of April and July 20 of 1977. During that time, OCSEAP observers made seven aerial surveys of the area, ADF&G management aerial observers made six aerial surveys, and private observers made another half dozen surveys of this area, as commercial interest in herring stocks in 1977 was high. It is because of these numerous surveys over a long time period, combined with the fact that surveyable quantities couldn't enter the system without being sighted, that the authors conjecture the herring simply didn't appear in 1977 or that they spawned unusually early. Although April spawning has not been noted in this area before, unusual climatological factors make early spawning a matter which must be considered. The winter of 1976-77 was the mildest on record in Alaska. In view of this, combined with the fact that Bering Sea herring normally enter their spawning grounds just after "ice out" (Barton, 1973), makes April spawning a theoretical alternative.

Herring were present on the spawning grounds for an entire month at the Meshik-Port Heiden sampling site in 1976, and for almost three weeks in Herendeen Bay. When the herring first arrived in Port Heiden, foreguts contained mysids, and herring were feeding daily. Soon after, foregut examination revealed that they ceased feeding, and gonad development changed from a gonad index of IV to VI in a matter of ten days.

Herring schools in the area of spawning were actively fed upon by numerous predators. Harbor seals, Phoca vitulina, were the most common form of mammalian predator, and caused schools to be in constant "amoeba-like" motion as they attempted to avoid the predations of the seal. Birds were another common predator. The species of bird feeding indicated the type of forage fish present, and some aerial observers were careful to note which type of bird was feeding on a school. Larids of the species Rissa tridactyla (Black Legged Kittiwake), Larus glaucescens, (Glaucous Winged Gull), and to a lesser frequency Larus canus, (Mew or Short Billed Gull), were most commonly seen in conjunction with forage fish, as well as terns, mainly Arctic Terns, Sterna paradisaea. Terns and kittiwakes are thought to be poor predators of spawning herring due to these bird's small size, while glaucous winged gulls and mew gulls may consume many herring during a single feeding. Often gulls were a good field indicator of schools for forage fish and observers were continually on alert for aggregations of feeding birds. Birds fed extensively on beds of herring spawn, and at low tides their activity was a good indicator of roe presence.

Needs for Further Study

The majority of data obtained during this study pertain to herring. As this species is receiving more and more pressure commercially, questions are being posed in regards to stock composition, range of distribution, migration routes, and what the potential effect petroleum related activities may have on all these things. These are subjects which we know very little about.

Fecundity studies have been often discussed as possible mean of population identification. Hodder (1972) also mentions that fecundity studies are important in "determining the relation between spawning potential and recruitment". To fully understand this parameter, fecundity sampling needs to be greatly increased to include a larger sample of the whole population. Fecundity methods need sophistication (see Appendix B); investment in automatic counting devices might be considered in order to eliminate this very tedious task (Boyer, et. al., 1967), and standardization of ovary collection must be accomplished. Field technicians must be fully aware of the importance of proper collection and preservation of fecundity samples. It is essential to have biological sampling start well in advance of the commercial fishing season and continue well beyond its completion to fully examine the developmental stages that the herring proceed through. Much work has been done on the identification of herring maturity stages, but little is know about how long it takes the fish to advance from one stage to another.

Capelin

General Life History

Capelin, Mallotus villosus is a widely distributed marine smelt which occurs in the boreal and arctic regions of the Atlantic and Pacific oceans. It is used extensively in the North Atlantic as a food and meal fish and harvests sometimes exceed a million metric tons (Jangaard, 1974). They spawn in the surf during spring on beaches and local residents harvest them at this time for domestic use. Male capelin, in both oceans, are larger and more robust in appearance than females, the former also having a distinctive "hairy" ridge along their lateral line, hence the specific name, villosus, which means hairy sided.

Discussion

Capelin are a short lived high productivity forage fish rarely exceeding three years of age. Most attain maturity at two years, and all mature by age three. The first year and a half of a capelin's life is spent in a larval state, as they do not metamorphose into the adult form until the middle of their second year of life (Templeman, 1948; Winters, 1966).

When adult males attain maturity, extreme body changes take place until in appearance they are quite different than females. These changes take place, it is supposed, immediately prior to maturity (Templeman, 1948). Prior to spawning, adults segregate by gender into schools (Pitt, 1958). At spawning capelin approach a suitable beach, spawning in a peculiar and characteristic way. Notes from direct observations by the junior author at Pillar Creek, Monashka Bay, near Kodiak City best describe the phenomenon.

> (My observations began) ...high tide was at approximately 2:00 a.m. and the weather was overcast, with an ambient temperature of approximately 50 degrees F; there was a slight offshore wind. Male capelin began to appear in the surf an hour before high tide and became increasingly numerous with each wave. The surf was checked carefully for females, but none appeared until the highest wave, which then carried in hordes of capelin. The spawning of these fish was just as spectacular as Templeman (1948) and Jeffers (1931) described and in my view is typical: with a wave of optimum height immediately at high tide, the fish swarmed onto the beach; it was obvious that female capelin definitely pair up with one or two males to spawn. Often with two males on each side of her, pinning her thusly. After spawning many hundreds of fish were stranded above the high tide mark, unable to return to sea. Of those sampled from stranded schools less than one in a hundred were female. Even in the sample of 100 fish taken in the surf for biological measurements I found only a dozen females, indicating to me that more females were yet to come onto the spawning grounds.

Templeman (1948) states that intense spawning occurs on beaches composed of pebbles between 0.5 to 1.5 cm in diameter. During field observations it was noted that sand on Pillar Beach, though not densely packed, was sandy in texture as opposed to the coarse pebble-like consistency indicated by Templeman. When spawning grounds were revisited a week later to obtain egg samples, none were found at the high tide mark where spawning occurred, but approximately two meters seaward from the high tide mark in coarser textured substrate, eggs were quite densely deposited.

Capelin spawning run timing is tied to a high tide phase in the mid-spring of the year, usually late May. What general mechanism enables the fish to detect the appropriate high tide phase is not known, yet certain meteorological factors concerning spawning do play a significant role. Offshore, rather than onshore, winds are an important factor for spawning capelin, to the point where they will not spawn if such a wind does not prevail. This had been noted by other observers (Winters, 1966; Campbell, 1973), and was also noted during this study. Anecdotal evidence from Aleut natives from the Meshik-Port Heiden area also supports this observation, as experienced elder natives told investigators that the direction of the wind decides if there will be a capelin run that night, i. e. that they require an offshore wind in which to spawn in appreciable numbers. Winters (1966) states that an offshore wind might allow the welling up of the colder water, thereby decreasing the water temperature and size of the waves. Proven reasons for the capelin's need of an offshore wind do not exist in the literature.

Deep water spawning, although not as frequent as beach spawning, has been reported in the Atlantic (Pitt, 1958; Templeman, 1948; Winters, 1966). During 1976-77 OCSEAP aerial surveys, capelin were often observed and photographed "digging" along nearshore subtidal areas and in fact this behavior became a field index to the presence of the species for aerial observers. It is not known whether this digging is in fact pre-spawning or spawning behavior, but presence of ripe male and female capelin with empty foreguts in the area suggests that this behavior is associated with spawning. Deep water spawning is yet an unknown factor in the spawning ecology of capelin along the north coast of the Alaska Peninsula.

After deposition, capelin eggs wash below the high tide zone where they had been deposited and become buried up to 150 to 230 mm in rock/gravel substrate averaging from 0.5 mm to 5 mm in diameter. Hatching takes place in widely varying time spans depending on temperature, although hatching times along the north coast of the Alaska Peninsula did not exceed two weeks, with 8 to 10 days being a minimum. After hatching the larva drift in the nearshore zone during the summer months and remain there until winter temperatures force them closer to the bottom (Templeman, 1948).

Windrows of dead capelin after spawning have often led observers to assume that capelin die after spawning, when this in fact is not the case. Evidence obtained by the senior author proves that capelin do live to spawn again as female capelin with retained and developing ova were found in mid-summer following spawning. These capelin had been captured in otter trawls. Recent findings support this result (Warner, and Dick, 1980, report in progress) as over a hundred post spawning males were found in commercial trawls, with full foreguts. Templeman (1948) reports occurrences of three "generations" of eggs in one female which indicates multiple spawning in Atlantic capelin.

Comparison of our fecundity at size results with Templeman's indicates Atlantic capelin to be 50 percent more fecund at size. After examining Templeman's findings on Grand Banks capelin one can see that the Atlantic capelin is both larger and much more fecund than those we examined from the Bering Sea. Other works on Atlantic capelin reiterate this fact (Hart and McHugh, 1944; Winters, 1966). The Atlantic capelin show a mean fecundity of 32 thousand eggs (Templeman, 1948) while our Bering Sea results showed a mean fecundity of 16 thousand ova per female capelin.

Needs for Further Study

Capelin research in the Pacific is sparse. Further research into the spawning activity and migration patterns of the capelin is needed especially in view of the growing commercial interest in capelin.

Capelin are known to occur in phenomenal numbers at spawning time, yet little is known about them in the Pacific after and before spawning occurs, especially as to overwintering demands and post-spawning migration.

Eulachon

General Life History

Eulachon are the most commercially important member of the smelt family in the North Pacific. They are an anadromous smelt, living entirely in the marine environment until spawning. Then they ascend rivers, sometimes many miles, to spawn. Their high body fat content gave them the name "candlefish", as the dried adults may be lighted and burned like a candle.

Eulachon have been harvested both commercially and domestically for over a hundred years. They are marketed under the trade name, "Columbia River Smelt".

Spawning Ecology and Spatial/Temporal Distribution

Eulachon appear to ascend river systems to spawn during their second through fourth year of life, and do repeat spawn in some instances. The ageing of eulachon from scales proved unsuccessful, as they absorb the outer margins of scales prior to spawning, rendering them unreadable (Smith and Saalfield, 1955; Warner and Dick, 1980 report in progress).

Little is known about the eulachon's high sea cycle. Presumably the eulachon in the contract area descend into the marine system immediately after hatching as they do in the Columbia and Fraser Rivers (Hart and McHugh, 1944; Smith and Saalfield, 1955). A sample of 178 immature eulachon were taken at sea by trawlers in the Kodiak area; results indicate that one year old eulachon were immature. Hart (1973) mentions that there is an indication of repeat spawning in eulachon, and 1979 observations in the Kodiak area reinforce this supposition, i.e. female gonads indicated they were formed subsequent to previous year's spawning.

Anecdotal information was gathered from local residents in the Alaska Peninsula region. This information further reinforced by documentation of historical runs of eulachon (see Appendix A) indicates that eulachon populations are highly cyclic, often reaching extraordinarily large populations then "crashing" until they reach a contrastingly low density.

It is because of these and other accounts, that the authors believe these fish are significantly more important as a forage fish than our catch results indicate. Indeed in some years they might conceivably be the most numerous forage fish species in the southeastern Bering Sea. Several accounts by local natives indicate that Bear River, Sandy River and the Meshik River had or have large runs of eulachon. It is important to note that even during years of low abundance, this species of forage fish has a documented high potential which strongly indicates a significant level as food fish.

Needs for Further Study

Other than a few catch statistics from British Columbia and the basic life history facts outlined in Hart's "Pacific Fishes of Canada", and Hart and McHugh (1944) "The Smelt of British Columbia", very little information was found on the occurrence, biology and distribution of this fish in the North Pacific, let alone the Bering Sea. In view of the total lack of current knowledge on this species further research is sorely needed.

Boreal Smelt

General Life History

Boreal smelt are anadromous, and they were originally classified by Linnaeus, and remain the archtype for the Osmeridae. Their distribution is circumpolar, rarely extending south of boreal regions. They ascend large river systems to overwinter, often migrating distances exceeding a thousand kilometers from the ocean, such as in the Yenisei River in Siberia (McPhail and Lindsey, 1970). At the end of each winter, stocks of boreal smelt spawn in tributary streams to large rivers immediately after "ice out" (Baird, 1967), then migrate into estuarine systems to feed until early autumn when they again ascend the river to overwinter (Hart, 1973).

Age

Boreal smelt in the Meshik-Port Heiden area were usually in their third year of life when sampled, although smelt from one to six years of age have been aged. Otoliths proved slow to process, and the confidence which both authors have in this method is low. Boreal smelt otoliths are large, in fact, so large they can be weighed with a balance and the figures then projected onto a Peterson graph where modes clearly indicate age groupings (Barton, Warner, and Shafford, 1977). By contrast, the mounting and reading of smelt scales proved rapid and easy. Some investigators of boreal smelt have mentioned they do not "put on" their first annulus until the end of their first year (MacKensie, 1958), yet when reading the scale it was observed that often the first annulus was directly in the nucleus of the scale or in contrast to this, six to eleven circuli distant from the nucleus. It was this feature which caused a discrepancy in the age values of the two readers. This obvious variance in the locus of the first annulus led the authors to investigate the scale topography of boreal smelt in depth, much in the manner that was used on capelin by Russian investigators (Prokhorov, 1968). Several references on this species indicated it has been known to have "double" runs on the Atlantic coast (Baird, 1967). The authors hypothesized that boreal smelt at Meshik-Port Heiden could be spawning twice a year, the same individuals not spawning more than once in a single year. In short, there might be an "iceout" spawning run and a fall or late summer spawning run. Since late spawners would have a very brief growth period after hatching prior to their first winter, the annulus would logically be centered or absent on the scale. In the case of spring or "iceout" spawners, these fish would have 90 to 100 days of growth prior to their first winter.

Spawning Ecology and Spatial/Temporal Distribution

The residents of the Bristol Bay area harvest this species through the ice when it is present in large river systems; it is used by both natives and non-natives as a subsistence/sports fish. Since boreal smelt are present in large numbers near the mouths of the Naknek, Egegik and Meshik Rivers, it is assumed to be of critical importance as a forage fish. Forty three percent of all forage fish caught at the Port Heiden test site in 1976 were boreal smelt, and they comprised 70% of the total catch at that site in 1977. Even during years of good herring abundance (such as in 1976) boreal smelt are the most numerous forage fish at this site. The authors have observed that boreal smelt seem to be most successful in river systems which have an expansive estuarine system at their mouth, in contrast to rivers which empty directly into the ocean with little or no estuarine tidal systems (i.e. Bear River, Sandy River).

It is hypothesized that boreal smelt, although migrating out into the marine system, stay within the estuarine system and perhaps only intermittently subject themselves to the rigors of the high seas predation. The reason the authors lean towards this hypothesis is that in 1976 during the cruise of the M/V PAT SAN MARIE, very few boreal smelt were caught in mid-summer (Bartlett, personal communication). At one time, it was thought that boreal smelt were present in large numbers in the southeast Bering Sea as investigators had given anecdotal evidence of their presence in trawl catches. Upon detailed examination, these fish were found to be juvenile eulachon, which are commonly confused with boreal smelt by professionals and laymen alike*. Hence, the authors wish to identify boreal smelt as being a species of the stuarine habitat.

Boreal smelt do not mature until their second or third year, and the numbers of recovering smelt which survive spawning indicates they may live to spawn twice, or even three times. In 1976, most of the smelt were either spawning or had spawned, while in 1977 almost 90% were immature. This is due to the fact that 1976 sampling activities began prior to or during the onset of "iceout" spawning, whereas in 1977, due to the mildness of the winter, "iceout" occurred in early March of the year. Since sampling activities did not begin until late May, the spawners had migrated into the open estuary and were not present in large numbers at the Meshik-Port Heiden test site. Boreal smelt in 1976 were found feeding while in a spawn ready condition. As the season progressed, this condition occurred less frequently until all fish found were "spent". This indicates that, even at iceout, boreal smelt do not spawn all at once, but do so gradually over a period of time.

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*Juvenile eulachon appear much different than mature fish of this species.

Needs for Further Study

Boreal smelt are a numerous species of which little is known. We have determined the possibility of there being a second summer run in the Meshik River system. An attempt was made in October of 1977 to document the occurrence of a second spawning run of smelt. Due to the extreme cold water conditions (1° to 2° C) in this system this fishing effort yielded only a single boreal smelt in three days. In 1976, a similar attempt was made at exactly the same time of year, and one hundred boreal smelt were caught while water temperatures of 6° - 8° C prevailed at Port Heiden. Boreal smelt were found to be in recovering spent* condition which indicates only that they had spawned previously, but giving no indication of when their spawning took place.

Perhaps the best approach would be to continue the spring field season far enough into the summer (perhaps into August) to afford sampling these fish over a greater time span. An earlier start of the field season would yield greater numbers of boreal smelt before their migration into the estuary to spawn. The temporal demands of boreal smelt appear dependent on iceout in the river system which they spawn, yet to how great an extent is not known. In any case, the time of their occurrence in the bays would definitely vary from year to year due to the varying severities of winter conditions.

Another aspect which needs continued documentation is the rate and amount of circuli laid down on scales prior to the first annulus. Extensive research was done on this subject in 1977 and it was from this work that the possibility of a second run of boreal smelt was hypothesized.

Aerial Surveys

Fewer schools of forage fish were seen along the north coast of the Alaska Peninsula in 1977 than 1976 when 649 schools of forage fish were observed. At Port Moller-Herendeen Bay no spawning herring were observed in 1977. Aerial observations along all areas failed to sight any windrows of dead capelin, an occurrence which was frequent in 1976.

Forage fish form dense ball shaped aggregations for protection against numerous predators, hence they are easily spotted from the air, but highly unpredictable to catch. Frequently the senior author observed large schools of spawning and pre-spawning herring from the air that were present in the immediate area of a surface camp, but none were captured there. During daylight activities, herring schools were observed passing unharmed under monofilament gill nets. On a single occasion, a school of herring in excess of 30 meters in diameter passed through a 25 meter long gill net without a single fish being captured. In short, zero fishing results were not indicative of the presence or absence of fish in the area being sampled. Ground crews in 1976 had difficulty catching herring. The primary tactic to ensure catches was to place experienced crews in areas where schools were sighted. When poor results were encountered by ground crews, aerial surveys were an expeditious method by which to reaffirm fishing results. Aerial surveys are the best means by which extensive sections of coastline may be surveyed in a very short period of time.

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*This being Stage VIII of the Gonad Maturity Index - See Appendix.

Needs for Further Study

The primary problem with aerial survey data in both 1976 and 1977 was the inability to positively identify the species composition of schools seen without surface verification. Consequently, subjective methods had to be utilized: 1) observation of fish carcasses on the beach (in the case of capelin), and 2) careful notation of the habitat type where schools were observed. (Herring require rocky shores with sudden rising escarpment, whereas capelin require open beaches with sandy/rocky substrate.) Further studies must be aimed at clarifying this problem. Use of acoustic and airborne laser methods for species identification should be explored.

Water Temperature And Salinity

Water temperatures are variable in some nearshore areas of the study area. As can be seen, extreme fluctuations occur within a close time period. This was especially the case in the Meshik-Port Heiden area. Port Heiden Bay is shallow and subject to extreme tidal variations, causing thorough mixing twice daily. Water temperatures in Togiak Bay were more stable than those in Meshik-Port Heiden because of the deeper water and less brackish conditions.

Herring first appeared on the spawning grounds at Port Heiden Bay in 1976 when the temperature was 6° C. In 1977, herring appeared in the Togiak region when the temperature was also approximately 6°. Onset of colder temperatures can affect forage fish movement and migration. In the fall of 1976 OCSEAP investigators caught over a hundred specimens in Port Heiden Bay, whereas in 1977 - precisely one year later - only a single specimen was captured. This was probably a direct function of water temperature, as in 1976, fall water temperatures were between 7° and 8° C, while in 1977 they ranged from one to two degrees.

Salinities taken for this study were determined by the use of a sodium chloride hydrometer. Although this instrument is portable and easy to use, it's accuracy is limited. Essentially, it reliably determines if the water sample is fresh, brackish, or of full sea water strength. Salinities were erratic at Meshik Beach and greatly affected by the magnitude of the outflow from the Meshik River and the daily tidal variations. In Togiak, salinities always remained nearly at full sea water strength throughout the 1977 spawning season. Catches from all areas didn't readily appear to be affected by variances or non-variances in salinity.

Miscellaneous Species

The Meshik-Port Heiden sampling site displayed, by far, a wider variety and abundance of non-forage fish species than did the Herendeen Bay site. This area is rich in potential and utilized finfish and invertebrate resources. In the early summer and late spring it is the location of a king salmon fishery, harvested by local residents. These king salmon ascend the Meshik River to spawn and are followed in June by a sockeye run in the Meshik River which supports a set net fishery. In the fall of the year a smaller salmon fishery exists in the area, yet besides these finfish, results indicate an impressive array and abundance of other species of finfish, the most abundant being char, Salvelinus sp.. Commercial quantities of anadromous char are resident throughout the Port Heiden-Port Moller area. They were the most numerous non-forage fish species and they were caught in large quantities by commercial fishing activities. Large starry flounders (over 40 cm) were caught by the hundreds in subsistence and commercial set nets in the Port Heiden area during June, July and August of 1977.

Few non-forage fish species were caught in Herendeen Bay. It is hypothesized that the lack of an extensive estuarine system in this immediate area accounts for this lack of non-forage fish species. Herendeen Bay, as opposed to the muddy waters of Port Heiden, is a clear salt water bay.

The Meshik-Port Heiden sampling site was located on a beach which was rich in invertebrates of potential commercial importance, and some (such as isopods) that aren't. This beach offers excellent habitat for soft shell clams of the genus Mya. The clam density at this beach averages approximately 25 to 40 "shows" per square meter. Field workers have frequently noted the strong aroma of sulfur dioxide emanating from the beach substrate, and numerous dead clams were found.

Large quanitities of crangonid shrimp and mysids populate Port Heiden Bay and natives frequently harvest them with small mesh seines. A hundred to a hundred and fifty kilogram catches of shrimp harvested in this manner have been witnessed.

Needs for Further Study

Although evidence from other disciplines indicates the presence of large quantities of sand lance (Sanger, 1977; Dick, personal communication) only three specimens have been captured in the two years of research by this agency in the Bering Sea. Trumble (1973) included quite a comprehensive summary of what is known of this species in the North Pacific Ocean in his Masters' Thesis on underutilized fisheries in Eastern North Pacific. OCS researchers in the Cook Inlet/Kodiak area have been successful in catching these fish with beach seines and surface tow nets (Blackburn, 1980; Warner and Dick 1980). So little is known about sand lance in our current study area that any research effort concerning it would be beneficial. They have been found to be important food for sea birds (Sanger and Baird, 1977) and for fur seals along the Aleutian Islands (Trumble, 1973). Therefore, the ecological importance of the fish in the food chain is justification for further research on the basic life history of sand lance in the Bering Sea.

Overall Discussion

To better understand forage fish, additional research must be completed. During the past two years this R.U. has raised many more questions than it has answered.

The possible impact of oil related activities on forage fish cannot be predicted with any accuracy since the effects of crude oil and/or refined oil products are virtually unknown in regards to Pacific herring, capelin, boreal smelt and eulachon. In addition to the actual products/byproduct itself, oil exploration and drilling activity has the potential of becoming extensive in the contract area and its effects upon forage fish are unknown at this time. Because of the speculative nature of this subject, therefore, the authors urge great caution be taken in any developmental aspect in the Bering Sea.

CONCLUSIONS

1. Capelin are a major forage fish in the Bering Sea nearshore area, in some years reproducing en masse along the beaches to the extent where windrows of trapped capelin .5 to 1.3 meters in height may be seen along miles of open beaches.
2. Capelin spawn offshore and onshore in the Bering Sea, not limiting themselves to the intertidal area.
3. Capelin in the study area are only half as fecund as those in the Atlantic.
4. Capelin occur on their spawning grounds for periods exceeding two weeks.
5. Surf smelt occur in the Bering Sea.
6. Boreal smelt are the dominant forage fish in the Meshik/Port Heiden estuary, being active there between May and July.
7. Boreal smelt show two patterns of scale topography which indicate variances in spawning and/or habitat patterns within the same population of fish.
8. Capelin and boreal smelt are able to spawn more than once.
9. Herring age composition in the Bering Sea area contained two strong year classes during 1977 studies.
10. Herring utilize Port Heiden for spawning.
11. More forage fish schools were seen north of Cape Constantine by aerial observers than on the north coast of the Alaska Peninsula.

12. Aerial surveys are able to determine relative abundances of forage fish, though species composition of sighted schools cannot be determined.

13. Water temperature and salinity changed considerably during brief time periods at Meshik-Port Heiden, and forage fish spawning took place during, after and before these changes.

APPENDIX A

AN UNUSUAL SMELT RUN AND ITS POSSIBLE EFFECTS ON RED SALMON ESCAPEMENTS INTO THE BEAR RIVER SYSTEM IN 1959

The Bear River red salmon system received an unusual and heavy spawning population of what was determined to be eulachon smelt in 1959. The Bear River spawning system consists of the headwaters of the lake, the lake shore, and the river which flows into the Bering Sea.

The timing of the smelt run coincided with the expected red salmon run throughout the month of July. Red salmon failed to ascend the river despite their abundance off the mouth of the river and in the offshore waters.

The vast size of the smelt run, its timing in conjunction with the red salmon run, and the subsequent demise of the spawning smelt could have resulted in an avoidance reaction by red salmon to enter the river in usual numbers, and also could have produced a temporary pollution condition in the stream.

The first aerial survey of Bear River was made on July 7th. The survey revealed the presence of an abnormal number of fish in the stream commencing from the juncture of the Milk and Bear Rivers to the entrance on the Bering Sea, a distance of approximately ten miles.

A second aerial survey of Bear River was completed on July 10th. The fish population in the stream had increased considerable since the previous survey on July 7th. It was estimated that the ten mile concentration numbered from two to five million fish or more. A low level flight revealed many fresh carcasses along the stream banks and also indicated the fish were probably a species of smelt.

A Super Cub was chartered from Sand Point on July 11th, and a landing was made on one of the numerous gravel bars in the river. It was a simple matter to reach in among the mass of fish to obtain specimens for indentification of the species of smelt. The specimens were identified as eulachon smelt, and several samples were frozen for shipment to Juneau for corroberation of the field indentification. Carcasses had increased considerable, although fresh smelt were still entering the river. A landing was also effected at the mouth of the river the same day. The dead carcasses were approximately eight to ten inches deep along the entire shore, and literally hundreds of thousands were lying along the banks of the stream.

The stream guard stationed at the mouth of the river stated the run had commenced about a week before the first survey which would place the entrance date at approximately the first week in July.

Appendix A

A third survey on July 15th revealed the run had peaked and was terminated. The banks of the stream were covered with dead smelt, but particularly in the lower sections of the river.

The stream guard reported the water had become badly polluted from the decaying carcasses, and also stated his drinking water was so badly contaminated that it was unfit for drinking purposes even after having been boiled. Water purification tablets were of little benefit.

Several fishermen reported at a later date that the ocean bottom in the offshore waters was covered with decaying smelt carcasses, and that the water was quite odiferous in the general area, and also that their nets were continually fouled up with the glue-like mass of decayed flesh.

The phenomenal size of the smelt run was discussed with old-time local residents, cannery personnel and perennial fishermen of the area. Everybody expressed surprise over the development in the Bear River, and could not offer any knowledge of smelt runs in past years. It may be possible that smelt runs did exist in Bear River in the past, but in such small numbers that they were unobserved.

APPENDIX B

FECUNDITY PROCEDURES

1. Take gonads in prespawning condition, i.e. no further along in maturity than Stage V; "Gonads fill body cavity. Eggs large, round; some transparent. Ovaries yellowish, testes milkwhite. Eggs and sperm do not flow; but sperm can be extruded by pressure". This stage passes into Stage VI which is when both "eggs and sperm flow freely". If ovaries are taken in Stage VI, there is too much of a chance of getting partially spawned out fish, thus erroneous egg counts. Another precaution is if there is doubt as to what stages the ovaries are in, check the posterior portion of the skein for membrane degeneration; if the membrane is intact, take the skein.

2. Preserve immediately in Gilson's Fluid[1] in plastic or glass jars, NOT Whirl-a-pak bags. The bags have been used in the past and have proven to leak. Leaking Gilson's fluid can be a serious problem!

3. Before beginning egg counts, decant Gilson's fluid and place in Lugol's Solution[2] for 24 hours. This will remove mercury from sample and decrease chances for allergic reaction to anyone working in and around the eggs.

4. Wash under running water or shake in jar until eggs are well separated. Strain through a wire mesh sieve, (careful of the size diameter opening!) lay out on paper toweling in petri dishes to drain adequately.

5. Place petri dishes of eggs in laboratory oven @ 60° C for 12 to 18 hours until eggs are entirely dry. Do not let them get dark brown and brittle.

6. Place samples in envelopes; seal.

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[1] Gilson's Fluid (Mueller, 1972): 88 mls H 0; 10.0 mls 95% ethanol (must be ethanol); 2.0 gr mercuric choloride; 0.4 ml glacial acetic acid; 1.8 m concentrated nitric acid. This solution is very good for breaking up membranes and is used widely for ovary preservation. It is, however, extremely toxic and corrosive. It should be used with caution. In handling preserved specimens, plastic gloves should be worn as some people find themselves extremely allergic to the mercury. It will attack metal, therefore, it should be stored only in glass or plastic containers. Mueller suggests that specimens be placed in Lugol's Solution (see footnote #2) for 24 hours to remove the mercuric chloride and then be transferred to water wash.

[2] Lugol's Solution: 1 gr potassium iodide, 0.5 gr iodine. Dissolve in 3 mls of H 0, then dilute to 50 mls.

7. Count out three subsamples for 250 eggs each, and weigh on a Mettler analytical balance to nearest .1 mg. Weigh whole sample to nearest .1 mg; calculate mean weight of 250 egg samples and figure fecundity by direct proportion.

8. A test run of ten subsamples per one individual should be done to show reliability. If two counters are involved, each should do a test run. This gives an indication of the reliability of your counts.

Some Special Notes and Precautions:

1. A grid system could be used for counting, such as a piece of heavy graph paper laid in the counting dish to avoid duplication and possibly ease eye strain.

2. It is possible that a dissecting scope or a very fine quality magnifying glass on a stand could be used to look through while counting the eggs.

3. Paper cupcake cups were used to hold each subsample as well as whole samples of dried eggs in the weighing process. Eggs and cup are weighed first; eggs dumped into whole sample cup and then paper weighed. Care should be taken in dumping eggs to avoid spillage and to be sure all eggs are evacuated before paper weight is recorded.

4. This author highly recommends a break in routine so that counters do not become fatigued. For example: Egg weights done for two days, calculation one day, then back to egg counting. For this year's egg counts it took two persons a full seven days of steady counting and weighing to complete the process. (This was to do 161 fecundities.) As can be seen, this is a very tedious and lengthy process, yet if your results are to be reliable, care should be taken to not push oneself too greatly during this stage of the game. Eyestrain is inevitable for either the counter or the weigher (using a Mettler analytical balance) and headaches may develop. Obviously efficiency and data reliability decrease with physical strain.

5. Mettler analytical balance is a delicate scientific instrument. One should be trained in its use by an experienced person. Ideally, it should be used in an enclosed room where people are not constantly coming and going. (So delicate is the mechanism that a person walking across the floor in its immediate vicinity can alter the reading!) Care should be taken to rezero balance frequently during a day's weighing procedures and to keep the inside compartment clean at all times.

APPENDIX C

Characteristics utilized to determine maturation stages of herring and assign maturity index numbers, Cape Sarichef to Smokey Point: April - September, 1976 - 1977.[1]

Maturity Index	Key Characteristics
I	Virgin herring. Gonads very small, threadlike, 2-3 mm broad. Ovaries wine red. Testes whitish or grey brown.
II	Virgin herring with small sexual organs. The height of ovaries and testes about 3-8 mm. Eggs not visible to naked eye but can be seen with magnifying glass. Ovaries a bright red color; testes a reddish grey color.
III	Gonads occupying about half of the ventral cavity. Breadth of sexual organs between 1 and 2 cm. Eggs small but can be distinguished with the naked eye. Ovaries orange; testes reddish grey or greyish.
IV	Gonads almost as long as body cavity. Eggs larger varying in size, opaque. Ovaries orange or pale yellow; testes whitish.
V	Gonads fill body cavity. Eggs large, round; some transparent. Ovaries yellowish, testes milkwhite. Eggs and sperm do not flow, but sperm can be extruded by pressure.
VI	Ripe gonads; eggs transparent; testes white; eggs and sperm flow freely.
VII	Spent herring. Gonads baggy and bloodshot. Ovaries empty or containing only a few residual eggs. Testes may contain remains of sperm.
VIII	Recovering spents. Ovaries and testes firm and larger than virgin herring in Stage II. Eggs not visible to naked eye. Walls of gonads striated; blood vessels prominent. Gonads wine red color. (This stage passes into Stage III).

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[1] From in house works; Data Collection Procedures for Herring, by Stan Moberly, no year.

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EFFECTS OF OIL CONTAMINATION IN THE SEA OTTER, *ENHYDRA LUTRIS*

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I. SUMMARY OF OBJECTIVES, CONCLUSIONS AND IMPLICATION WITH RESPECT TO OCS OIL AND GAS DEVELOPMENT

A. The objective of this study was to measure effects of crude oil contamination on sea otters through studies on the changes in the animal's physiology and behavior before and after contact with oil. A second objective was to attempt to rehabilitate the otters after crude oil contamination.

B. Conclusions:

The study has shown that small amounts of crude oil contamination have large effects on the metabolic rate of sea otters. Light oiling of approximately 25% of the animal's pelt surface area resulted in a 1.4X increase in metabolic rate while immersed in water at 15°C. Furthermore, when the oil was removed by detergent, the animal's metabolic rate increase 2.1X while immersed in water at 15°C. Of the three animals studied, two contracted pneumonia and one died. Studies upon free ranging sea otters have established that under certain conditions, sea otters can sustain low levels of oil contamination when 20% or less of the body surface is oiled.

C. Implications:

Any contact with oil at any time of year would have a profound influence on the health of individual sea otters through increases in the animal's thermal conductance and the subsequent increase in metabolic rate. It is probable that death may follow from pneumonia or hypothermia depending upon the amount of the animal's fur fouled. Rehabilitation of oil-fouled sea otters would be very costly requiring holding facilities to keep the animals for at least two weeks. Even if adequate facilities were available, the success rate of rehabilitating oil-fouled sea otters is likely to be rather low.

II. INTRODUCTION

A. General Nature and Scope of Study

This project represents an extension of studies that began with the investigations of diving and feeding behavior, and thermoregulatory effects of oil pollution in fur seals (Kooyman et al., 1976a). In this species, which relies on fur for insulation against cold sea water, the effects of oiling were profound. It is thought, but not measured, that sea otters are even more dependent on their fur for insulation.

It was predicted that oil would have an impact on sea otters through increases in their maintenance costs due to increased heat loss in air and in water as a result of pelage contamination. The magnitude of this increase will also vary between neonates and adults. External oil may also impair their diving and feeding abilities. Kenyon (1974) noted that malnutrition was common in contaminated fur seals. Finally, we anticipated direct metabolic effects of ingested oil. This research continued physiological research on sea otters, and included an analysis and evaluation of other studies on effects of oil pollution on sea otters. The results will provide an answer to such questions as well as help in providing basic information on the general aspects of respiration in all marine mammals.

Furthermore, determination of the characteristics of diving and measurements of the energetics involved will help to provide general information on the energy requirements of the various marine mammals and their different modes of propulsion.

B. Specific Objectives:

1. Energy requirements of normal sea otters at various water temperatures.
2. Energy requirements of sea otters after oiling.
3. Appropriate procedures for rehabilitating oiled sea otters.
4. Energy requirements of washed sea otters and time required for complete recovery.
5. At-sea behavior of sea otters.
6. At-sea behavior after oiling.

Information of this kind will provide a data base from which the assessment of any kind of oil contamination or other activity which may alter the nature of the otter's food sources can be derived. In addition, relative to oil contamination, the difficulties and costs of protecting from oiling and the rehabilitating of oiled otters can be estimated.

C. Relevance to problems of petroleum development:

The sea otter is a conspicuous faunal element of many nearshore communities in Alaska and California where offshore oil development and transportation is either underway or planned. As fur bearers, they are among the marine mammals most likely to be affected by oil. Furthermore, the California sea otter population has been determined to be a threatened population by the Department of the Interior due primarily to the threat of an offshore oil spill. By measuring the physiological responses of individual animals to surface contamination, the present research effort provides basic information from which the impact of an oil spill within the sea otter's range can be predicted.

III. CURRENT STATE OF KNOWLEDGE

The effects of oil spills on marine mammals have been reviewed recently by Geraci and Smith (1977), and by Davis and Anderson (1976). From both these sources, it appears that hair-bearing marine mammals (either adult or immature) are not usually killed by simple surface contact with oil. However, fur-bearing marine mammals (either young or adult) are much more susceptible, and may be killed by such contact.

Our previous study has shown that small amounts of crude oil have a large effect on thermal conductance of fur-bearing pelts, and no effect on nonfur-bearing pelts. In living fur seals light oiling of approximately 30% of the pelt surface area resulted in a 1.5X increase in metabolic rate while immersed in water of various temperatures. Furthermore, this effect lasted at least two weeks.

Preliminary research by Siniff Johnson and Williams (1977) indicates that crude oil contamination of live sea otters results in death under certain circumstances. However, their investigation was of a limited nature and no conclusions can be made from their study. Metabolic rates of immersed sea otters have been measured previously (Morrison, Rosenmann and Estes, 1975; Iverson and Krog, 1973) and are useful only in comparisons with control animals used in the present study.

IV STUDY AREA

Laboratory experiments were conducted at the Physiological Research Laboratory, Scripps Institution of Oceanography, University of California at San Diego, California. Captive sea otters were collected from Monterey, California with the assistance of the California Department of Fish and Game.

Field studies were conducted in Constantine Harbor, Hinchinbrook Island, Prince William Sound, Alaska (146° 38' north latitude; 60° 22' west longitude) during July 7-22, 1978 and June 20-August 17, 1979.

V. SOURCES, METHODS AND RATIONALE OF DATA COLLECTION

A. Laboratory studies

Five female sea otters, *Enhydra lutris* weighing 12 to 20.9 kg were used. They were captured near Pacific Grove, California by employees of the California Department of Fish and Game utilizing the diver held sea otter capture device. (Wild & Ames, 1974). Soon after capture, the animals were transported by rented aircraft to a holding tank at the Physiological Research Laboratory, Scripps Institution of Oceanography, San Diego, California. The time of capture to release in the Scripps pool was about 5 hours. Animals were maintained for periods of 2 months to 2 years in a holding tank that was 6.1 m wide, 12.2 m long, 3 m deep. The tank was filled with sea water to a depth of 1.2 m. Fresh sea water flowed through the tank at approximately 110 ℓ/min. Water temperature within the holding tank varied with changes in the ambient ocean water. Summer temperature averaged 19°C and the winter temperature averaged 16°C. Five times a day , the sea otters were fed a variety of food items including commercially obtained frozen clam, *Spisula solidissima*, squid *Loligo opalescens*, rock crabs, *Cancer spp.*, abalone trimmings, *Haliotus spp.*, and locally collected sea urchins, *Strongylocentrotus spp.* Upon completion of the study, the otters were transferred to Sea World, San Diego, California.

Metabolic Rates during Immersion - Prior to each metabolic analysis, the sea otters were fasted for 12 hours. The otters were weighed wet to within 50 g on a platform beam balance just prior to each experiment. Soon after weighing, all animals were placed in a metabolic chamber specially designed for these studies. The chamber, which was 151 cm long 84 cm wide, 84 cm deep, held about 1400 liters of fresh water. It was constructed of styrofoam sheets, 9.5 cm thick, which were covered with a wood veneer and fiber glassed for strength. Within the lid was a 30 x 60 cm lucite dome fastened tightly over a neoprene gasket. The box was filled with fresh water to 2 cm into the dome. Opposed ports in the lower portion of the dome functioned as intake and exhaust for air drawn through this air space. The dome was covered with black plastic to prevent the otter from being disturbed by the personnel conducting the experiment.

The rate of air flow was measured with a Wright respirometer accurate to within 1% at flow rates used. Humidity was determined with a dial hygrometer, and barometric pressure was measured with an aneroid barometer checked against a mercury barometer. Air and water temperature of the box were monitored with an accuracy of 0.1°C with a Bailey bat 4 coupled to a digital voltmeter. The thermocouple probes were placed at the mouth of the air intake and on the upper portion of the chamber wall about 2 cm below the water level. Water was slowly, constantly and uniformly stirred in the box by means of a series of outlet and inlet manifolds. Water temperature usually varied less than 0.5°C.

A sample of the exhaust from the dome was drawn continuously through a complex of 3 glass "U" tubes filled with drierite, barlyme (a CO_2 absorber), and drierite, in that order, before entering the sensing cell of an Applied Electrochemistry Industries oxygen analyzer (AEI). In those experiments with oiled or washed animals, a "U" tube of 4-12 mesh activated charcoal preceded the first drierite "U" tube to absorb all oil fumes.

The AEI 0_2 analyzer signal was recorded continuously on a 25 cm chart recorder adjusted to record from 19-21% full scale. At 60-minute intervals, the inlet air sample was checked and the instrument's reference cell adjusted if it had drifted. At this time, the analyzer was calibrated by flushing the sensing cell with room air, which was presumed to be 20.93% 0_2.

The recorded curves of 0_2 concentration were smoothed by eye and the difference in 0_2 concentration from the intake and exhaust were determined every minute. The averages for 60 minute intervals were collated. Appropriate factors for correction of gas volumes to STPD were incorporated into a computer program, and oxygen consumption rates (V_{0_2}) were calcualted using the following equation (Depocas & Hart, 1957):

$$\dot{V}_{0_2} = \frac{\dot{V}_{1_{stpd}} \, (F_{1_{0_2}} \cdot F'_{E_{0_2}})}{(1 - F'_{E_{0_2}})} \qquad (1)$$

where

$\dot{V}_{0_2}$ = oxygen consumption (liter/min)

$\dot{V}_{1_{stpd}}$ = Incoming air flow

$F_{1_{0_2}}$ = Fraction of oxygen in dry ambient air (.2093)

$F'_{E_{0_2}}$ = Fraction of 0_2 in CO_2 and water free exhaust

Surface Area Measurements - The surface area of 5 fresh pelts were measured by tracing the pelt, paws, tail and flippers on an acetate sheet. The outline was then transferred to tracing vellum and the outlines were cut out and weighed.

The determined surface area to weight ratio of the tracing material was then used to calculate surface area. Total surface area of one pelt was measured by planimetry and by weighing; both procedures agreed to within 1%.

The surface area of the otters' appendages (tail, paws, flippers) were measured separately by planimetry. The surface areas of the hind flippers were measured while both the flippers were expanded and while they were closed. A surface area constant a was derived from these measurements and it was used in the equation (Iverson & Krog 1973):

$$S = a\,W^{2/3} \qquad (2)$$

where:

S = total surface area in square meters $(\text{meters})^2$
a = constant derived from surface area measured from the 5 otter pelts.
W = weight of animals (kg)

in order to calculate the surface area of the experimental otters.

Deep Core and Subcutaneous Temperature Measurements - Deep body temperatures were obtained by inserting an encapsulated radio transmitter down the esophagus of anesthetized animals. Two deep core temperature transmitter systems were utilized. One system used a procedure in which temperature was modulated into a varying pulsed interval ratio. The receiver then demodulated this signal to an analog voltage which was then measured on an oscilloscope. The other method used a design of Mattison and Seeley (1974). In this unit, the transmitted pulse rate varied with temperature and was determined by counting the number of pulses per minute timed with a stopwatch. The accuracy of these systems was accurate to ± 0.1°C.

Subcutaneous temperatures were determined with small (approx. 2 cm x 1 cm) surgically implanted AM radio transmitters designed by Mckay (1970). The transmitted signal and temperature measurement was similar in procedure to that of Mattison and Seeley (1974).

Experiments with Oiled and Washed Otters - In order to measure the loss of insulation due to crude oil contamination and due to the washing of the fur to remove the oil, four experiments were conducted on 3 of the 5 otters (once each on otters 2 and 4 and twice on otter 1).

Sea otters were oiled under light anesthesia. This was accomplished by placing the whole animal in a nylon bag and flowing air containing 5% vaporized halothane into the bag until the animal was unconscious. After the induction, the otter was removed from the bag and the otter's head was placed in a plastic cylinder through which gas at a known concentration of halothane was flowing. Gas flow rate and concentration were maintained with a Bird MK 5 respiratory and fluotec vaporizer.

Prudhoe Bay crude oil was brushed over a prescribed area of the animal's back. This process took no more than 15 minutes. After oiling, the animals were permitted to swim in a clean pool of sea water for 30 minutes before the metabolic test.

Upon completion of three of the metabolic tests, the animals were anesthetized and cleaned with Amberlux detergent. Cleaning took 30-45 minutes after which the animals were returned to the holding tanks and fed. Twenty-four hours later, a second metabolic test was run to measure the effect of washing. In the first oiling experiment, otter 1 was washed twice, once immediately after the post-oiling metabolic measurement and again the next day. In the final experiment, oil was left on otter 5 for 8 days prior to washing. Post-oiling metabolic measurements were made twice during this period on days 1 and 6. This oil was removed with a shampoo and conditioner, formulated by Redkin Laboratories, Los Angeles, California.

B. Studies on Free-ranging Sea Otters:

In order to study the behavioral response of oil contaminated sea otters, a field camp was set up on the south shore of Constantine Harbor, Hinchinbrook Island, Prince William Sound, Alaska (146°38' north latitude; 60° 22' west longitude). The behavior patterns of oiled and control (unoiled) animals were monitored by radio telemetry. Activity patterns were monitored by a remote telemetry station located on a hillside overlooking the study area at an elevation of 1000 ft. A Telonics (Mesa, Arizona) TR-2, 150-152 mhz biomedical telemetry receiver with a Telonics TS-1 Scanner/Processor was interfaced to an Esterline Angus 20 channel event recorder via a specially designed pulse detector. This detector converted the audio output of the receiver to a voltage which deflected the appropriate event recorder pen when a signal from a otter radio transmitter was received. Each channel was scanned at least once every 30 seconds. Radio telemetry collars were constructed so as to float upon release from the animal. The collars were constructed of an inner core consisting of closed cell urethane foam which was sealed with a thick coating of silicone rubber, to prevent the collar from becoming saturated with water. The collar base was then covered with a brilliantorange polyester jacket onto which a Telonics 150 mhz band B-5 tracking transmitter was attached. The collars were fastened around the otters'neck with velcro strips which went through magnesium-stainless steel buckles. The buckles were designed to corrode away,releasing the collars after 4 days for collars with depth recorders attached or 14 days for collars with tracking tansmitters alone.

The foraging depth of sea otters was to be studied with specially designed maximum-depth recorders attached to the telemetry collars of 4 sea otters. These recorders digitally recorded the depth of the last 64 dives. Upon release, the collars were to be recovered and the data retrieved from the depth recorders.

Sea otters were captured using "tangle nets". The tangle nets were modified 9" mesh gill-nets which floated on the surface of the water. They did not possess lead lines and were 150 ft long and 10 ft deep. One end of the surface line was anchored via a float to the bottom so that the net could swing with the tide. Two nets were deployed in areas frequented by sea otters. While foraging or swimming, otters would wander into or under the nets and become entangled. Once the otters were entangled, we would come along side the net in a boat, haul the otter aboard and anesthetize it with a portable halothane gas anesthesia apparatus. While under anesthesia, radio collars and flippers tags were attached. Oiled animals were treated the same as controls except that 10-30 mls of Prudhoe bay crude oil was brushed onto the

the fur covering no more than 30% of the dorsal surface. The otters were maintained on board until they recovered from anesthesia, whereupon they were released into the water.

Activity and behavior patterns of the oiled and control sea otters were monitored by the received radio signal patterns. Upon submersion, the telemetry collar signal was lost to the salt water and was not received by our telemetry receiver. Active periods were typified by a broken signal pattern and rafting periods (resting on the surface), a constant signal pattern. During daylight hours, observations were made to validate the recorded telemetry patterns with the animals' behavior. Observations of the grooming pattern of oiled animals were also made at this time.

Sea otter population surveys were also conducted during the field seasons. During July, 1978 approximately 2/3 of Prince William Sound was surveyed utilizing a Bell Jet Ranger 200. During August, 1979, a UH-1H helicopter was used to survey the coastline between Cordova, Valdez and the Columbia Glacier due to poor weather and helicopter mechanical problems. Two people counted and a third person recorded the otter sightings on the appropriate charts.

VI. RESULTS

A. Laboratory Studies

A total of 67 control metabolic experiments, totalling over 402 hr metabolic measurements were made on five female sea otters immersed in water at 5,10,15,20,25 and 30°C. The mean resting or standard metabolic rate measured in these animals at all water temperatures was 12.0 ± 1.2 ml O_2/kg-min or 4.0 ± 0.4 Watts/kg (Table 1).

A significant difference was not measured in resting $\dot{V}_{O_2}$ from 5° to 30°C; however, a significant increase in both average and active $\dot{V}_{O_2}$ was observed as the water temperature decreased ($P < .001$, Analysis of Variance, Figure 1). The mean amount of the time spent active appeared to increase as the water temperature decreased (Figure 3).

Surface area measurements for the five sea otters are presented in Table 2. The total body surface area can be calculated from equation (2) where a = 0.087. The appendages, including the tail, accounted for 21.4% of the the total body surface (hind flippers 12.4%, tail 4.8%, paws 4.2%). The hind flippers can be expanded or closed, resulting in a 25% reduction in flipper surface area or a 3% reduction in the animals' total surface area.

Mean body temperature for three animals was 37.2°C (R = 36.7 - 38.1). There was a 12°C difference between the subcutaneous temperatures in a control otter immersed in 15°C and 5°C water. Upon oiling, the overall body temperature beneath the oiled area dropped 5-10°C (Table 3). Once the otter was washed or if oil was left on for several days, the subcutaneous temperature underneath the oiled area increased, approaching body temperature (Table 3). Figure 3 displays the variability of body temperature in otter 5 which was washed and immersed in 15°C water. Changes in T_b correspond with changes in T_{sq} in otter 5 after washing. The lowest body temperature are coincident with the start of activity and an increased heat production (Figure 3).

Oiling experiments were conducted on three sea otters, once on otter 3 and 5, and twice on otter 1 (Figure 4). The subcutaneous and core body temperatures for the four oiling experiments can be seen in Table 2. The average metabolic rate of otter 1 increased 22% after oiling 16% of her body surface with 38 mls of crude oil. After washing, her metabolic rate increased 102% above the control rate. Eight days after oiling, her metabolic rate returned to normal. In the second experiment, 13% of otter 1's surface was oiled with 60 mls of crude oil. After oiling, there was a 69% increase in her normal metabolic rate. After washing, her metabolic rate was 125% above normal and was still 33% higher than normal after 14 days. In the third oiling experiment, 25% of the body surface of otter 3 was oiled with 60 mls of oil. Her average metabolic rate increased 36% after oiling and 111% after washing. In the fourth oiling experiment, 23% of otter 5's body surface was oiled with 35 mls of crude oil. After oiling, her average metabolic rate increased 35%. In this experiment, oil remained on the otter for 8 days. The average metabolic rate was 63% greater than the control 1 day after oiling and was 98% higher than control 6 days later. Washing the otter with a shampoo and conditioner supplied by Redkin Laboratories (Los Angeles, California) resulted in a metabolic rate that was 87% higher than control, three days later, the metabolic rate was 75% higher than control and 8 days post-washing, it was still 75% higher than control. After washing, the sea otters' furs appeared wet and the animals were observed shivering. The lower subcutaneous temperatures observed in oiled pelt areas suggests an increased thermal conductance of oiled fur.

After the first oiling, otter 1 recovered in our normal holding facility. However, after the second and third oiling experiments, the washed otters were incapable of completely grooming their fur into its normal condition. Forty-eight hours after washing, otter 1 had groomed only her upper torso and the fur on her lower abdomen was totally wetted. During this time, she was constantly shivering. The water temperature of our holding tank was 16.7°C. Due to constant shivering and the inability of otter 1 to groom herself properly, she was removed from the holding tank and placed in a small tub filled with 25-30°C water. The tank was left filled for three hours and then drained and left empty for 1-2 hours and then filled again. This procedure was carried out for 24 hours. After the 24 hour period, the otter had managed to successfully groom her entire body and was returned to our regular holding tank where she continued to groom normally and was not observed to shiver again.

Two weeks after oiling otter 1, we took a routine blood sample and measured a white blood cell count of 16,500 cells/mm^3. The normal white count is around 8,000 cells/mm^3. Otter 1 was then transferred to Sea World for veterinary care, where they diagnosed pneumonia. Otter 1 recovered after treatment with antibiotics. When she was returned to the Physiological Research Laboratory 100 days after oiling, her fur was matted and her metabolic rate was 71% greater than normal.

After washing otter 3, we put her into a tank of warm water (26-30°C). However, unlike otter 1, she would not groom. Over the next six day period, we put her into cold water then warm water and then left her dry. She did attempt to do some grooming but was unsuccessful in restoring the air layer into her fur. On the sixth day after oiling, we transferred her to Sea World, where they diagnosed initial signs of pneumonia. Five days later, she died (11 days post-oiling). A necropsy was conducted and revealed gross inflammation of the lung from acute pneumonia. No problems were encountered with otter 5 after washing. She appeared to be able to withstand the thermal stress without grossly noticeable ill effects. However, otter 5's fur retained a slick appearance long after the washing. This was probably due to the inability of the new shampoo to completely remove the crude oil. The otter was transferred to Sea World and when observed four months later otter 5's fur appeared normal.

The holding tank temperature on the day of the oiling experiments was : 1st experiment otter 1, 20.2°C, 2nd experiment, otter 1, 16.4°C, otter 3, 16.1°C and otter 5,20°C.

B. Field Studies

A total of 31 sea otters were captured using the tangle net capture device during the 1979 field season. Of these 31 animals, 3 drowned during capture, 7 were oiled, 15 were used as controls, and 4 were used as depth recorder controls. Two were a mother and pup pair which had to be anesthesized to be removed from the net and were then released untagged and uncollared. Temple cattle ear sized tags were used to identify individual animals after the radio collars released (Table 4).

We collected consistent telemetry data from 4 of the 19 control animals and 3 of the 7 oiled. Some of the telemetered animals removed their radio collars immediately upon release and others left the area or transmitted intermittant signals. Intermittent two-to-five-hour direct observational periods were correlated with the strip chart records and used for interpretation of these data. Three types of behavior were identified in the strip chart records; rafting, floating quietly on back with very infrequent activity such as one or two rolls, or rubbing of paws or face; low level activity, low intensity grooming with rolling or slow swimming; high level activity, vigorous grooming or foraging. These three behaviors were summarized as 1) percent of time spent in each activity for each animal, 2) mean percent time spent in each activity for control versus oiled animals, 3) mean time spent in a rafting bout per animal and for control versus oiled, and 4) mean number of rafting bouts per day per animal.

The radio-collared otters were monitored for anywhere from one to twenty-one days. The daily activity patterns of six animals (four control and two oiled) are presented in Tables 5-7. Oiled animals spent a mean of 20% of their time in low level activity, 20% in high level activity, and 60% in rafting. Oiled animals spent a mean of 113 min in a rafting bout and rafting bouts took place an average of 9 times per 24 hour period. Control animals spent a mean of 115 min in a rafting bout and rafting bouts took place an average of 8 times per 24 hour period. However, significant differences were not observed between the activity of oiled and control sea otters.

The location of two of the oiled otters (tag #671,672) which moved out of range of our telemetry receiving station were monitored with a hand-held telemetry receiver from a boat. The seven oiled otters were oiled with 10-30 mls of crude oil covering 1-10% of their body surface. Two of the oiled otters left the study area within the first 12-24 hrs of capture (tag #s 665,669). One left the study area within 48 hours of capture (#662).

One oiled otter died 18 hrs after capture. Upon necropsy, severe peritonitis was determined to be the cause of death. We believe the animal was ill when captured and that the oil was not the direct cause of death. Otters 664 and 672 had 2.3% and 10% of their body surface oiled with 15 mls and 30 mls of crude oil, respectively. They both were alive and in apparently good condition when the study ended. Otter 664 was monitored for 43 days and otter 672 for 20 days. We were able to visually observe the behavior of otter 664 on 4 occasions for a total of 14 hours. During these observations, otter 664 spent 63% of her total grooming time grooming her oiled fur. After 21 days, she had substantially reduced the area of visibly oiled fur. Thirty days after oiling, a matted area of fur could still be seen.

Otter 671 had 8.1% of her dorsal surface oiled and appeared healthy 20 days after oiling. However, 26 days after capture and oiling, she was found dead. She apparently died from a large open neck abrasion caused from the tightness of the telemetry collar. The severity of the neck wound indicated to us that oil contamination was not the primary cause of dath.

The depth of six dives were recorded from otter 663, 5 of the dives were to 33 ft and 1 dive was to 52 ft.

The results of the population survey can be seen in Figures 5-9. Figures 5 shows the overall area surveyed. These figures are modified from Calkins, Pitcher & Schneider (1975).

VII. DISCUSSION

A. The mean standard metabolic rate in water of 12.0 ml O_2/kg-min is slightly different from the 11.2 ml O_2/kg-min in air and 14.2 ml O_2/kg-min in water reported for sea otters by Morrison et al. (1974). This oxygen consumption rate is 2.4 times the rate predicted for a terrestrial animal of equal size (Kleiber, 1975). It is 1.8 times more than other mustelids (Iverson, 1972) and is similar to the resting level of other marine mammals. Thermal adaptation to the marine environment seems to result in increased rates of metabolism in marine mammals (Hart & Irving, 1959; Morrison et al., 1974). It is possible that this increased metabolism is necessary to compensate for the greater thermal conductivity of water, which is 20-26 times greater than air (West, R.D., 1977). Hart and Irving (1959) found that the thermal conductance in harbor seals doubled upon immersion in water. For comparative purposes, we can calculate the thermal conductance across the pelt of the sea otters resting in water, if these conditions are met or assumed: 1. body temperature remains constant, 2. cutaneous and respiratory heat loss are insignificant when compared to overall heat flux, 3. The relationship of oxygen consumed to energy produced is 1 ml O_2/min-kg = 0.335 W/kg. Thus, we can calculate the total thermal conductance from the following expression:

$$C = \frac{M}{(T_{SQ} - T_A)S} \qquad (3)$$

where

T_{SQ} = subcutaneous temperature (°C)

T_A = ambient temperature (°C)

M = metabolic rate (Watts)

C = total thermal conductance (Watts/°C m^2)

S = surface area (m^2)

Utilizing the above equation, we calculated a mean thermal conductance of 5.9 $\pm$ 0.8 for the otters studied at 15°C. The sea otter, polar bear, adelie penguin and fur seal all have similar thermal conductances (Frisch et al., 1974, Kooyman et al., 1976b, Kooyman et al., 1977). Polar bear skin does not remain dry when immersed, but relies on a stagnant water layer for insulation (Frisch et al., 1974).

There is a disparity in the conductances between pelts and the coats of animals in both sea otters and fur seals. This difference is a result of the need to groom the fur frequently in order to prevent water penetration and restore dryness of the fur (Kooyman et al., 1977).

The sea otter's ability to adjust to varying water temperatures without increasing resting oxygen consumption is striking (Fig. 1). Since heat production equals heat loss, the sea otter must be employing mechanisms which reduce its heat loss or increase its overall total heat production or both. One mechanism for reducing heat loss without increasing oxygen consumption is by peripheral vasoconstriction. Peripheral vasoconstriction in sea otters was quite striking with a drop in subcutaneous temperature of 12°C in water between 5° and 15°C(Table 2). The thermal flux calculated at 5°C was 7 $w/m^2 °C$ and 5 $w/m^2 °C$ at 15°C. The small increase in thermal flux at 5°C was probably due to the 12°C drop in T_{SQ}.

Morrison et al. (1974) noted that sea otters immersed in 30°C water floated very low with their abdomen almost submerged. They postulated that the otters allowed water to infiltrate their fur decreasing the fur's conductance and the otter's buoyancy. We also observed our sea otters floating very low in the water at higher temperatures(30°C). However, we did not observe water infiltrating the fur. The texture and appearance of the fur remained the same before, during, and after the metabolic experiments. As the water temperature declined, the otters seemed to float higher in the water. Sea otters may vary their lung volume at different water temperatures to increase or decrease the body surface in contact with the water. This would alter the heat flux of the animal. This orientation in the water could partly explain the lack of increased O_2 consumption with decreasing water temperature. Increasing the amount of body surface in contact with the water at high water tempertures would increase heat flux. However, in nature, there may be a compromise, such that at high wind velocities and very low ambient air tempertures, heat loss could be lower with total immersion in cold sea water (Miller _et al._, 1975).

Sea otters seem to increase their overall heat production but not their resting metabolism. The level and duration of activity, such as grooming, increase as the water temperature declines. For example, we measured a significant increase in overall oxygen consumption below 20°C and a significant rise in the intensity of activity below 20°C (Fig. 1). The relative time spent active also increased at lower water temperatures (Fig. 2). Core body temperature varied over a 1.0°C range and it was correlated with active and quiet periods (Fig. 3). The metabolic heat produced during activity seems to be stored, which results in an increase in body temperature. In quiet periods the resting metabolism of the sea otter is normal which results in a gradual loss of heat and T_B declines.

Variations in core body tempertures of 1°C or more with activity have been recorded in other marine mammals, such as elephant seals (McGinnis and Southworth, 1971), spinner dolphins (Hampton and Whittow, 1976), monk seals (Ohata _et al._, 1972), and sea lions (Whittow _et al._, 1972).

The flipper surface relative to total body surface is smaller in sea otters than in other marine mammals and in the river otter _Lutra_ (Iversen and Krog, 1973; Tarasoff, 1974). A smaller surface area to mass relationship may be important to sea otters which apparently have not evolved the elaborate vascular counter-current exchange systems found in the flippers of other marine mammals (Tarasoff, 1974). The hind flipper temperatures in sea otters are high (32°C) and are usually greater than abdominal skin temperatures (18-25°C) (D. Costa, unpublished observation). The ability to expand the flipper surface by 25% may be an important mechanism for altering heat flow from the body during rest or activity. Sea otter flippers are highly vascularized (Tarasoff, 1974) and would act as an effective heat radiator. Prior to every rafting bout, sea otters groom their fur. The last areas of the body groomed are the flippers and paws (Loughlin, 1977). As the flippers, head and paws are groomed, they are licked dry by the otter; and while rafting, they remain out of the water (D. Costa, personal observation). This behavior would aid in heat retention during rafting when heat production is low and aid in heat loss during activity (while flippers are in contact with water) when heat production is high.

The changes in the sea otter's metabolic rate after oiling and washing parallel those previously observed in live fur seals and in pelts (Kooyman _et al._, 1976a). In both studies, the post-wash rate was twice that of the control. The fact that the fur becomes wet to the skin after washing suggests that the natural fur oils have been removed. If this is true, our data indicate that a minimum of 8 days is required for the natural fur oils to be replaced before the metabolic rate (therefore overall thermal conductance) return to control levels (fig. 4).

The difficulty encountered during recovery in two of the oiling and washing experiments correlates with the holding tank temperatures. In the first and fourth oiling experiments (otters 1 and 5), the tank temperature was above 20°C and the animals recovered without special handling. In the second and third oiling experiments (otters 1 and 3), the tank temperatures were around 16°C and both animals became ill and one died.

The high subcutaneous temperature in otters emphasizes the importance of the fur in maintaining a thermal gradient to reduce heat loss (Frisch *et al.*, 1974). The lower subcutaneous temperature under the oiled fur indicates that due to the loss of insulation, there is a localized reduction in peripheral circulation. The increased subcutaneous temperature below the washed fur and below fur left oiled is unexpected (Table 3). There may be a limit to how long and widespread the otter can maintain peripheral vasoconstriction.

Because of the high subcutaneous temperatures measured under washed fur, there must be a larger heat loss due to the thermal gradient between the water and the otter's skin.

We can calculate the thermal conductance across oiled and washed fur by utilizing a modification of equation 3:

$$\frac{(M_X - M_C)}{(T_{SQ} - T_A)S_X} + C_C = C_X \tag{4}$$

where

M_X = oiled or washed heat production (Watts)

M_C = control heat production (Watts)

S_X = surface area oiled (m^2)

T_{SQ} = subcutaneous temperature under oiled or washed area (°C)

T_A = ambient water temperature (15°C)

C_C = control thermal conductance calculated for each animal with equation 3 ($W/m^2\,°C$)

C_X = oiled or washed thermal conductance ($W/m^2\,°C$)

The changes in thermal conductance can be seen in Table 8. The surface area affected by washing was estimated to be twice the area oiled. These results are about the same as 27 $W/m^2\,°C$ for oiled and greater than the 21 $W/m^2\,°C$ on washed pelts measured by Kooyman *et al.* (1977).

Heat flux across the oiled fur is a function of the density of crude oil contaminating the fur (Fig. 10). Utilizing the mean oiled thermal conductance we can calculate the total heat production of an 18 kg otter entirely covered with crude oil to be 224 Watts or 3.1 times the resting level. This is close to the resting level we recorded of 2.6 times resting after otter 1 was washed. Under this condition, the fur offers little insulation and therefore seems to substantiate the validity of our oiled calculation. In this state, the otter was shivering violently, a sign of great thermal stress. It is unlikely that an otter completely oiled with crude oil while in a cold water environment of 15°C or less could survive. Note that the above calculation only represents the initial increase in thermal conductance. Several days after the initial oiling, the fur's conductance might increase as much as twofold, as in otter 5 (Table 5).

From the above, one might draw some useful conclusions about proper procedures in the treatment of oiled otters. It is apparent that washing can be quite destructive to the fur's insulating quality. The high conductance values measured after washing in otter 1 are probably an overestimate due to a greater area of fur being affected than was actually used to calculate conductance. In order to gain an estimate of the lower limit (and probably a more valid estimate) of thermal conductance after washing, we can assume all of the animal's fur was affected by washing. In this condition, we can use the animal's total surface area and equation 4 to get conductances of 15.3, 14.1, 14.8, and 15.1 W/m^2 C for the four experiments respectively. It is likely that the higher oxygen consumption measured after washing was not entirely due to the greater thermal conductance of the washed fur, but due to a marked increase in the overall surface area affected. Therefore, when planning to wash oil-fouled sea otters, it would seem prudent to clean only those areas which were soiled, thereby decreasing thermal stress to the animal.

B. The most striking results of our field studies are that the sea otters survived small amounts of oil contamination and that we did not measure a difference in their activity patterns. These results appear to contradict our laboratory studies. However, the amount of oil contaminating the wild sea otters was less than that contaminating the captive animals.

For comparison, we can estimate the increased thermal load experienced by the wild otters utilizing the data derived from the captive otter studies. The increased heat lost through the oiled fur can be calculated by rewriting equation 3:

$$M = (T_{SQ} - T_A)\ ^{\circ}C \cdot S \qquad (5)$$

where: T_{SQ} = subcutaneous temperature under the oiled fur (°C)

T_A = ambient water temperature (°C)

C = thermal conductance of oiled fur ($W/^{\circ}Cm^2$)

S = surface area oiled (m^2)

M = heat flux (Watts)

The thermal conductance of the oiled fur, C, can be derived from Figure 10 given the density (volume/area) of oil applied to the fur. The oiled subcutaneous temperature is assumed to be the same as in our captive animals since the ambient temperature of oiled captive animals and oiled wild otters were similar (15°C, 14°C respectively). The results of these calculations as well as the relative increase in the resting energy consumption (derived from resting energy consumption of captive sea otters) are presented in Table 9. The increase in resting heat production needed to offset the additional heat loss due to oiling was only 11-22% in otter 662,664 and 671. The increased heat production ranged from 22 to 39% in oiled captive sea otters (Table 1). The increased activity or other thermoregulatory mechanisms utilized to offset the heat loss due to oiling may not have been detectable with our activity telemetry system. However, we visually observed that otters 664 and 671 spent greater amounts of time grooming the oiled areas than the unoiled areas. We have no data for otter 672, who was expecred to have the greatest increase in energy production, because it wandered out of the range of our primary telemetry receiver.

Another complicating variable between captive and free-ranging sea otters is the added stress of captivity. Smith & Geraci (1975) found zero mortality in experimentally oiled harp seals in the wild, but observed high mortality in oiled captive harp seals. They attributed this difference in part to the increased stress of captivity. Captivity stress would be an important complication in any rehabilitation of oil-fouled sea otters.

Using the preceeding methodology, we can estimate the increased metabolism for an average wild Alaskan sea otter receiving differing amounts of oil over varying areas of fur. The effects of oil-fouling on an Alaskan sea otter weighing 28 kg (=mean weight of those captured) can be seen in Figure 11. Notice that when oil densities exceed 600 ml/m^2 covering 8% or more of the animal's body, the calculated increase in metabolism is greater than the highest level measured in captive sea otters under any condition.

These relationships plotted in Figure 6 represents a model, and sea otters may undergo a variety of thermoregulatory adjustments which this model does not accomodate. In addition, this model only predicts the initial changes in metabolism and we would expect the thermoregulatory stress to increase as the otter continues to groom the oil into its fur. This effect was observed in otter 5 who was oiled for 8 days (Figure 4). However, this model allows us to make these useful predictions about crude oil contamination of sea otters:
1. lighter oils which result in a thin coating of oil upon the animal's fur may not have as great an impact on sea otters as heavier oils which would result in a thicker, denser coating of oiling; 2. otters may be capable of thermoregulatory compensation when only a small percentage of their body surface is oil fouled.

C. Population Survey

Lensink (1962) estimated the 1960 Prince William Sound - Kayak Island sea otter population at 1000 to 1500 based on fixed-wing aircraft observations of 702 animals. According to Schneider (1971), this estimate may be in error by as much as 100%. The Alaska Department of Fish and Game (1973).

estimated the population at 5000, which is reasonable in light of the numbers of otters counted in the 1973-74 surveys: 2015 in June, 1973 compared to 1441 in March 1974 (Calkins, Pitcher, and Schneider, 1975), both were helicopter surveys. In July 1978, 2148 otters were counted when slightly less area was surveyed (Figure 5).

Hinchinbrook-Hawkins Islands

The number of sea otters in this census block was increased as a result of significant growth in localized areas. Hinchinbrook Island has several embayments and harbors in which sea otters are well established. Constantine Harbor is still densely populated, but it appears that carrying capacity has been reached and population growth has stabilized. On the other hand, the Port Etches area supports a much larger population than previously, perhaps as a result of overflow from Constantine Harbor (Figure 6).

The southeast side of Hinchinbrook was relatively bare compared to previous years, but the number of otters increased as the mouth of Orca Inlet was approached. The Inlet population as observed in March, 1974 appears to be maintaining but not rapidly expanding.

The otters which have migrated from the Knowles Head-Port Gravina area to the Sheep Bay area in the 1974 survey are still present, although only a pod of thirty animals was observed compared to 202 otters in 1974 (Pitcher, 1975).

Knight Island Area

There does not appear to have been a significant increase in the number of sea otters in the Knight Island area (Figure 7). A small population with a few pups was spotted around Eleanor Island. Around Ingot, Sphinx, and Disk Islands, only isolated, single otters were observed. The general pattern around Knight Island is one of small groups of otters wide-dispersed along the coast. Very few pups were present.

Montague Island

Like Hinchinbrook Island, there is a trend evident here toward population growth in restricted areas. The area around Graveyard Point and into Stockdale Harbor is now the home of a sizeable population with many pups (Figure 8). Port Chalmers inlet continues to support a dense population and numberous pups were present. Hanning Bay, Macleod Harbor, Jeanie Cove, and Patton Bay all support only small groups of otters at this time. Otter sightings were rare between Box Point and Zaikof Point, so that the east side of Montague Island remains basically unpopulated. Significant numbers of otters exist in both Zaikof and Rocky Bays.

Valdez Arm-Port Gravina Area

It is especially crucial to monitor the otter population in the Valdez Arm-Port Gravina area (Figure 9) since this is the site of heavy oil tanker traffic. By comparison between 1973-74 and recent survey results,

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a dramatic increase in sea otter numbers is apparent. One hundred seventy-seven animals were observed in this area in 1973, 73 in 1974, and 974 in 1978.

After his survey, Pitcher (1975) stated:" areas which appear to be acceptable habitat but are not presently supporting significant sea otter populations include Perry Island, Bligh Island and Galena Bay." A major trend in population recovery in this direction is evident from the data, as otters have spread to Goose Island, up into the entrance of Port Fidalgo, especially in Snug Corner Cove and Boulder Bay, and Bligh Island, all of which were nearly unpopulated less than five years ago. Further, range expansion was observed in 1979. Ninety-two sea otters were spotted in Jack Bay and 1 in Galena Bay, where none were observed in 1978.

It is evident from the data that the sea otter population in Prince William Sound has increased in size over the past five years, and the process appears to be continuing. Dense populations now exist in specific areas which previously did not support sea otters. Of major significance is the population growth and range extension occurring in the Valdez Arm-Port Gravina area, since these otters are moving into sites that are in direct contact with the oil traffic waterway. The sea otters in this area are especially susceptible to oil contamination, due to the nature of the small and narrow bays and ports. An oil spill could close the entrance to one of these bays or ports, making it impossible for the otters to leave without becoming oil fouled.

VIII CONCLUSIONS

The impact of crude oil contamination upon sea otters can be quite profound and subsequent rehabilitation of oil-fouled sea otters would be both difficult and costly.

Oil contamination results in a profound increase in the heat loss of sea otters - Field studies have shown that sea otters can survive low level oil contamination under certain circumstances. Field studies were conducted under optimal conditions, i.e. abundant food supplies and warm weather. In areas of marginal sea otter habitat or high sea otter population densities, where competition for food is high (such as Amchitika Island, where seasonal die-offs of juveniles have been reported (Kenyon,1969) sea otters may be more sensitive to oil contamination. Weather and environmental temperature will also influence sensitivity to oil contamination. Sea otters will be more sensitive to thermoregulatory stress where the sea water temperature is lower than in our study area (mean sea water surface temperature 14°C). Sea otters will be more sensitive during the winter when environmental temperatures are low;periods of sustained stormy weather (times when oil spills are more probable) will reduce the foraging ability of sea otters, also making sea otters more sensitive to oil contamination.

Damage to sea otter populations is more likely from large scale oil spills where there is a higher probablity of contaminating a large proportion of the animal's fur. Contamination of large areas of the animal's fur (greater than 30%) will most likely result in death. Sea otter's may be resilent to chronic low-level oil contamination, where the probability of large surface oil contamination of the fur is low. The probability of a sea otter making contact with oil also is less likely with chronic low-level oil spills. A large scale oil spill within an area populated by sea otters could result in oil fouling of most of the sea otter population and would most likely result in death for those animals oiled. However, if oil contamination does not directly foul sea otters, its effect on sea otter prey items must be considered. Sea otters must consume 20-30% of their body weight daily in prey. An oil spill which substantially reduced the number of available sea otter prey items would also result in a considerable die-off of sea otters. More continued research into the avoidance of oil spills and the effects of oil on sea otter prey and improved rehabilitation techniques are needed.

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Table 1. Mean resting oxygen consumption (mls O_2/kg-min) for each experimental period at various water temperature

	Water Temperture °C								
Otter	5	10	15	20	25	30	Oil	Post Oil	Wash
	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$	$\frac{ml\ O_2}{kg\text{-}min}$
1	14.4 17.7 11.14 13.2	12.0 10.8	12.0 13.7 9.7 10.4 11.9 12.7	12.4 14.2	12.9 12.9 14.4 11.7 12.0	12.7 10.4			
Exp.1							15.9		31.2
Exp.2							16.3		27.7
2	15.4	--	--	11.9 13.0	9.2	--	--	--	--
3	14.1 12.2	10.8 11.1	11.2 12.0 9.7	9.7 11.2 11.3	10.5 10.6	10.5 10.9	14.8		26.0
4	12.6 6.0	10.7 11.5	15.3 13.8 9.5 9.6	8.5 8.3	11.1 9.1 9.6	9.1 9.4			
5	7.1 16.0	15.2	11.4 13.8 13.2 12.8	10.1 12.0	11.9 13.7	13.0 11.5 12.0	13.5	15.5	28.8

Table 2 - Surface areas measured from the pelts of 5 sea otters

Sex	Body Weight kg	Hind Flippers Expanded m^2	Hind Flippers Closed m^2	Paws m^2	Tails m^2	Maximum total Surface area m^2
F	16.6	-	-	-	-	0.5
M	20.0	1.020	0.782	0.331	0.238	0.805
M	24.9	0.649	0.453	0.209	0.340	0.657
F	35.8	1.386	1.120	0.440	0.378	0.902
M	36.3	1.067	0.774	0.425	0.648	0.965

Table 3. Subcutaneous (T_{sq}) and deep core body temperature (T_B) in °C in control, oiled washed, 1 day post-washed and 1 day post-oiled animals. All temperatures were measured from otters immersed in 15°C water unless noted otherwise. Numbers below temperature are one standard deviation, numbers in parentheses equals the sample size.

* Temperature was recorded under normal fur of a partially oiled otter.

Animal	Control		Oiled		1 day Post-oiled	Washed		Post-wash
	T_{sq}	T_B	T_{sq}	T_B	T_{sq}	T_{sq}	T_B	T_{sq}
Otter 1 Exp. 1	36.4 ±0.3 (4)	36.7 (2)	26.2 ±0.7 (19)	37.3 ±0.2 (12)	-	34.2 ±0.3 (7)	-	35.8 0.4 (15)
Exp. 2	32.3* ±1.1 (15)	-	26.0 ±0.6 (14)	-	-	-	-	-
Otter 1 at 15°C	36.6 0 (4)	37.6 0 (4)	-	-	-	-	-	-
Otter 1 at 5°C	24.3 - 	37.7 0 2 37.2	-	-	-	-	-	-
Otter 2 at 15°C	-	36.7 ±0.22 (21)	-	-	-	-	-	-
Otter 3	-	-	28.3 ±0.3 (21)	38 1 - (2)	-	34.5 ±2.2 (19)	-	-
Otter 4	-	37.9 (3)						
Otter 5	-	-	24.6 ±0.4 (37)	37.6 ±0.2 (38)	33.7 ±0.3 (4)	35.0 ±1.3 (34)	36.8 ±0.5 (24)	
$\bar{X}$	35.1	37.3	26.3	37.7	33.7	34.6	36.8	35.8

Table 4. Summary of the tag number, weight, sex, date of capture and treatment of the 31 sea otters captured during the 1979 field season.

Otter Tag #	Sex	Weight kg	Treatment	Date
652	F	23.1	Control	6/28/79
653	F	21.3	Control	6/28/79
654	F	24.9	Control	6/29/79
655	F	22.7	Control	6/29/79
656	F	21.8	Control	6/30/79
657	F	23.1	Control	6/30/79
658	F	22.2	Control	6/30/79
659	F	22.2	Control	6/30/79
NT(no tag)	F	21.8	Control	6/30/79
660	M	38.6	Control	6/30/79
661	F	22.7	Control	6/30/79
662	M	40.8	Oiled with 10 mls over a 10cm x 15cm area	7/7/79
663	M	36.7	Control depth recorder	7/7/79
664	F	26.3	Oiled 16 mls 10x18cm area	7/7/79
665	M	34.9	Oiled 18mls over a 11x15cm area	7/13/79
666	M	41.7	Control	7/13/79
--	F	24.0	Drowned in net	6/30/79
667	M	35.8	Oiled 30 mls four dead next day of peritonitis	7/17/79
668	M	37.2	Control with depth recorder	7/17/79
669	M	33.6	Oiled with 20 ml over 30x19 cm area	7/19/79
670	M	35.4	Control	7/20/79
671	F	27.7	Oiled with 20 ml over a 26x25 cm area	7/20/79
--	M	37.6	Drowned in net	7/22/79
--	M	36.3	Drowned in net	7/22/79
--	F with pup	--	Released mother as soon as seen in net	7/20/79
672	F	21.3	Oiled with 30 mls over a 26x26 cm area	7/30/79
673	F	20.4	Control with depth recorder attached	8/1/79
674	M	30.1	Control	8/1/79
675	F	16.3	Control with depth recorder attached	8/1/79
--	M	20.0	found dead on shoreline died natural causes	7/22/79

Table 5. Activity patterns over a 21 day period of a free-ranging oiled sea otter (otter 664)

Date	Low Level		High Level		Rafting		Unknown
	Min	%	Min	%	Min	%	Min
7/8	354	25	227	16	859	59	
9	77	7	93	9	910	84	360
10	109	11	184	19	667	70	480
11	512	36	261	18	667	46	
12	324	23	304	22	787	55	25
13	280	19	305	21	854	60	
14	238	16	329	23	873	61	
15	275	20	302	21	823	59	40
16	257	18	314	22	869	60	
17	298	21	250	17	892	62	
18	383	27	319	23	712	50	26
19	230	16	255	18	955	66	
20	309	21	247	17	884	62	
21	150	10	222	15	1068	75	
22	261	18	217	15	962	67	
23	57	4	366	26	1006	70	11
24	160	11	470	33	810	56	
25	377	26	317	22	746	52	
26	285	20	303	22	824	58	28
27	416	29	372	26	652	45	
28	266	20	273	20	796	60	105

Table 6. Mean activity patterns for six otters (otters 664, NT, 663, 675,673, 662) and for oiled versus control animals, in percent time.

Otter	Low Level	High Level	Rafted	n(days)
664-oil	19	20	61	21
NT	15	30	55	11
663	13	23	64	5
675	34	10	56	3
673	18	18	66	12
662-oil	13	13	74	2
Oil	16	17	67	23
Control	20	20	60	31

Table 7. Mean time spent in a rafting bout and mean number of rafting bouts per 24 hour period for six otters

Otter	$\bar{X}$ time/rafting bout	$\bar{X}$ number rafting bouts/24hr
664-oil	114 min.	8
NT	124	7
663	88	10
675	110	7
673	139	7
662-oil	112	10
Oil	113	9
Control	115	8

Table 8. Thermal conductance calculated for the control; oiled and washed fur.

Otter	Oiled density ml/cm^2	Control flux W/°C m^2	Oiled flux W/°C m^2	Oiled 6 days flux W/°C m^2	Washed flux W/°C m^2
1 Exp 1	0.05	5.65	27	-	39
1 Exp 2	0.08	4.58	57	-	47
3	0.04	5.75	19	-	23
5	0.03	6.03	10	37	28
$\bar{X}$	0.05	5.50	28	-	34

Table 9. Shows the resting metabolism, the area oiled and the oiled fur heat flux used to calculate the increased heat production resulting from oiling.

Otter #	Resting Metabolism W	Area Oiled M^2	Heat flux Oiled fur $\frac{W}{M^2 {}^{\circ}C}$	Oiled Metabolism W	Increased in Metabolism %
662	164	.0150	13	44	12
664	106	.0180	23		22
671	111	.0650	12		11
672	86	.0676	30		34

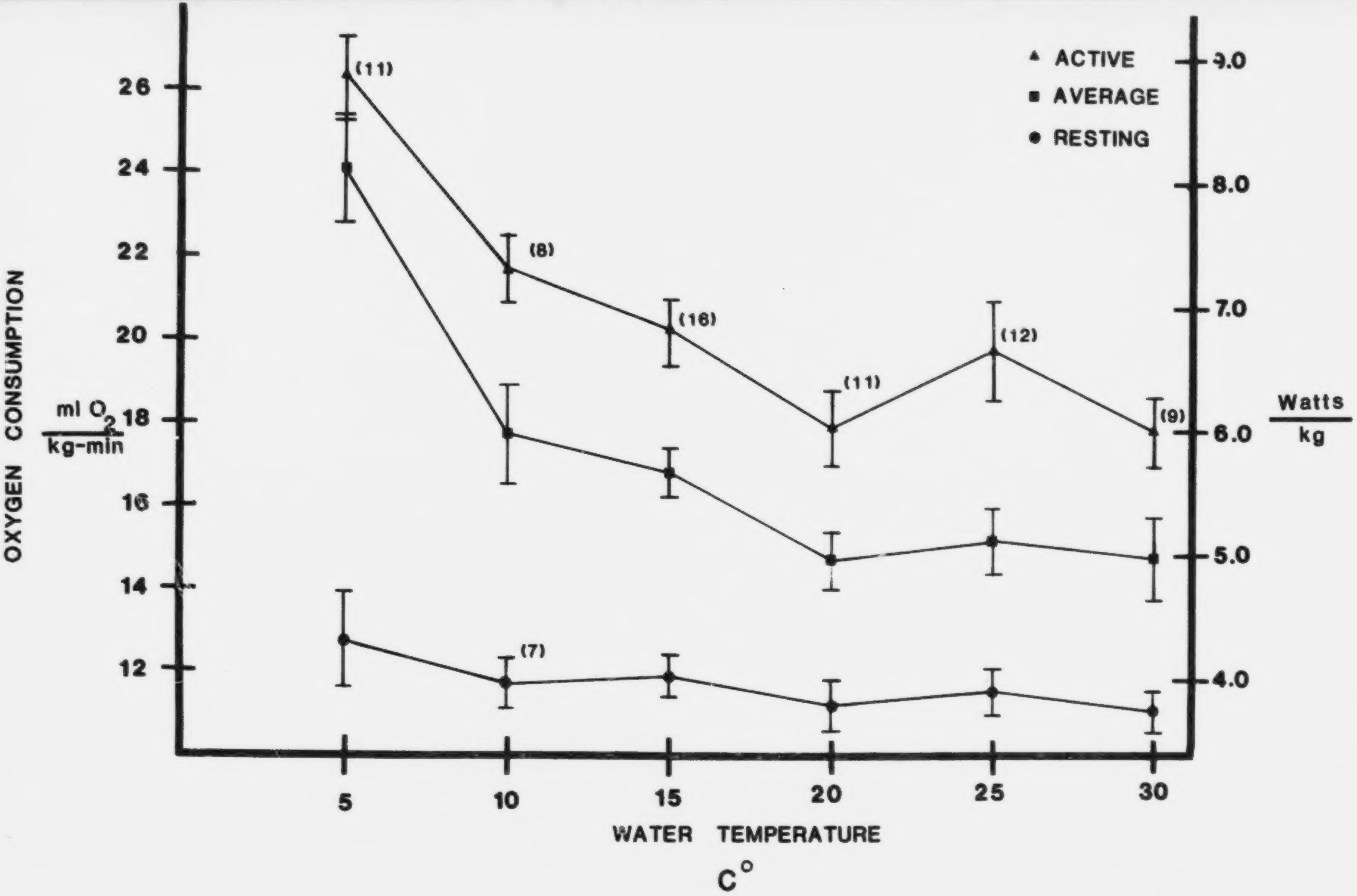

Figure 1. Resting, average and active oxygen consumption are plotted against chamber water temperature for the 5 sea otters studied. 1 standard error is plotted for each point. Each point represents the mean of the means for each experiment run. The mean number of experiments used to determined the points on the figure are given in parentheses.

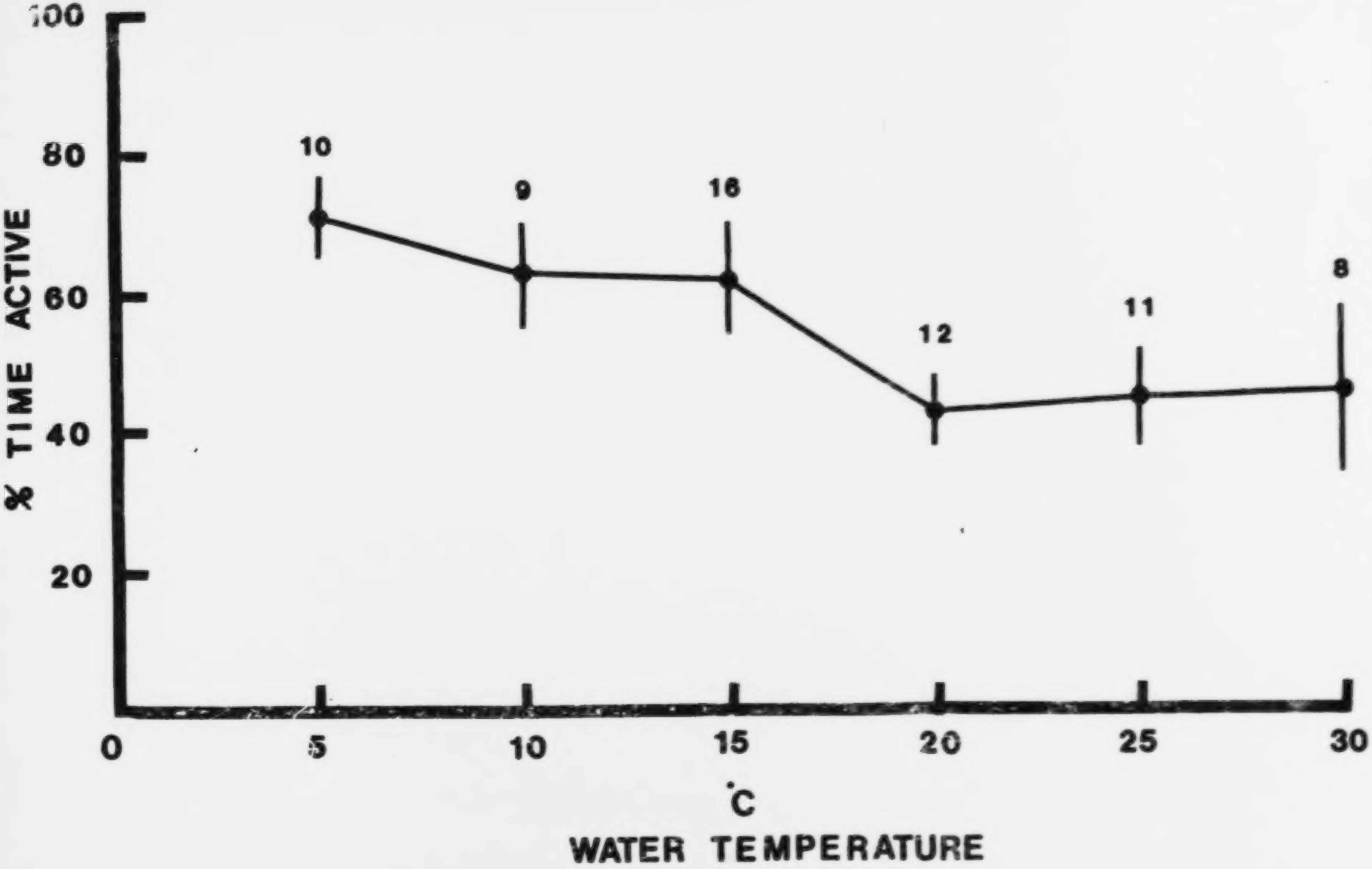

Figure 2. The mean percent time spent active during the six hour metabolic experiments is plotted against water temperature. The numbers above each value correspond to the number of 6-hour metabolic experiments at each water temperature. The bars represent one standard error.

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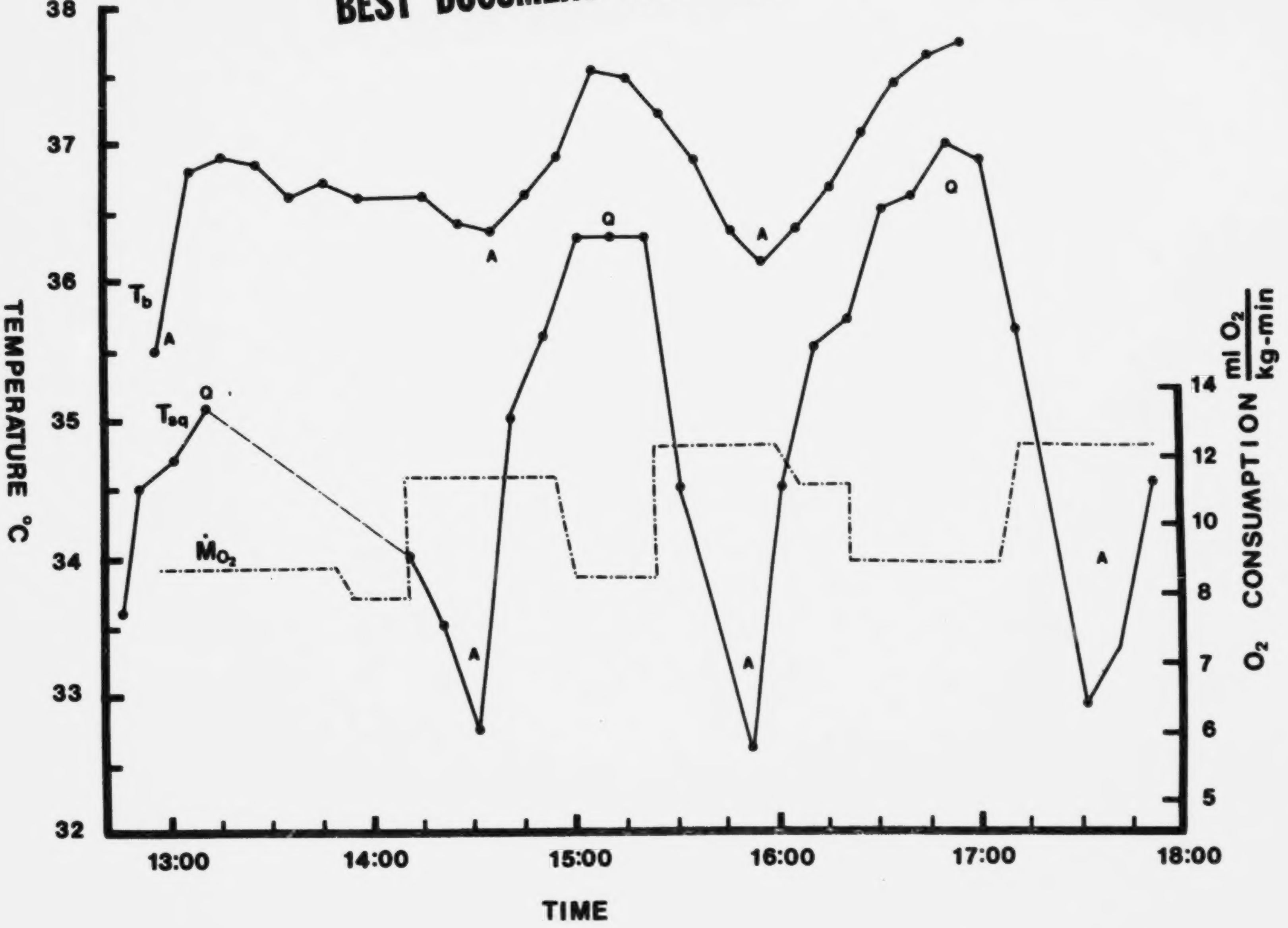

Figure 3. The T_B (body temperature) T_{SQ} (subcutaneous temperature) and oxygen consumption ($\dot{M}O_2$) are plotted against time measured on otter 5 after being washed. The A represents active periods and Q represents quiet periods. The dashed line in T_{SQ} represents a break in the data.

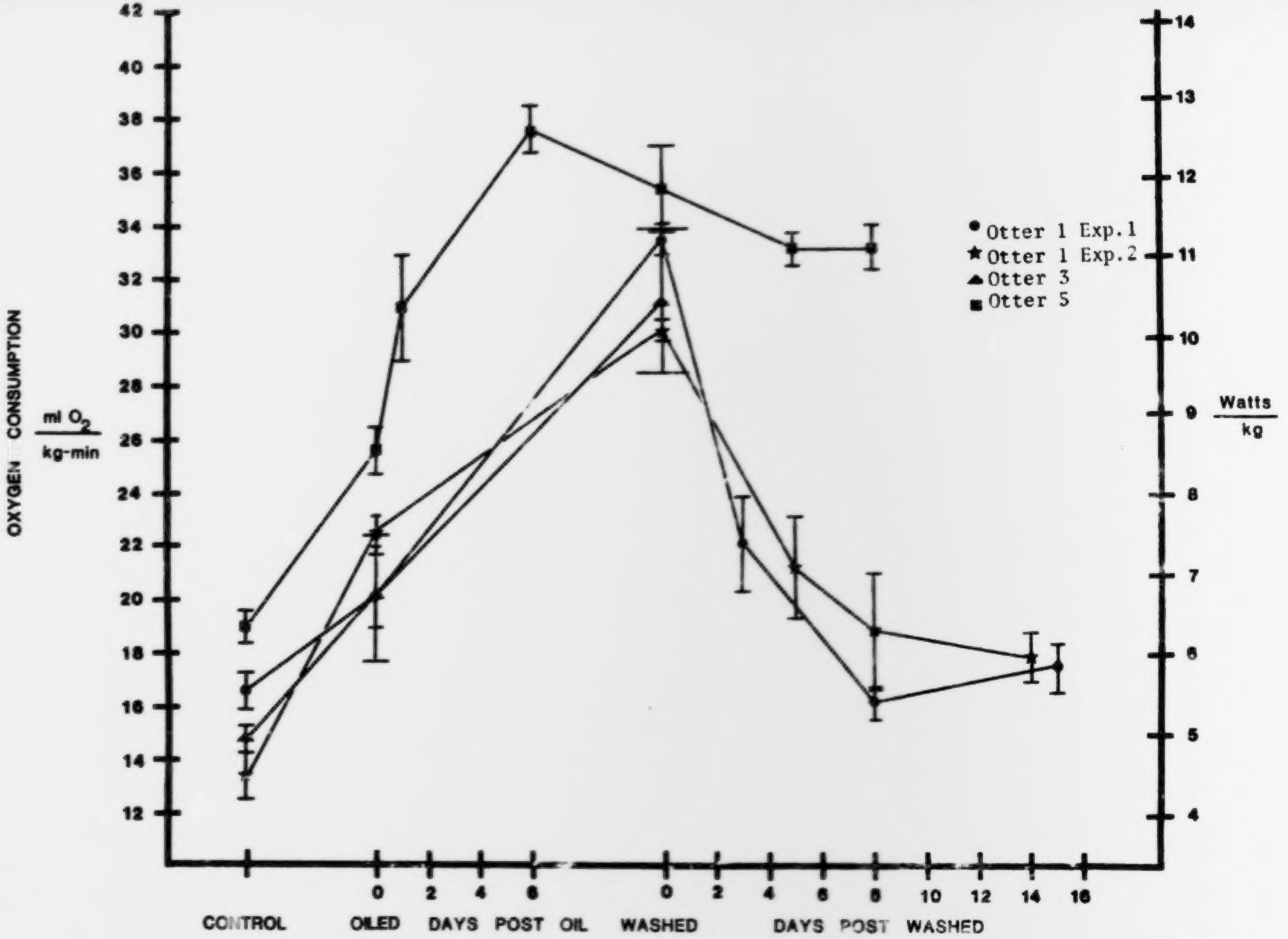

Figure 4. Mean average oxygen consumption for control, oiled and washed animals are displayed. Days post oiled only refers to treatment of 5 where the oil was left on 8 days prior to oiling. Otherwise, all animals were washed 8 hrs after oiling except for 5. 1 standard error is plotted as bars around the mean. The number of measurements for control runs was 18 otter 1, exp 1;12 otter 1, exp 2; 16 otter 2; 24 otter 5. For oiling and washed runs 6 otter 1 exp 1; 3 otter 1, exp. 2; 6 otter 3; 6 otter 5.

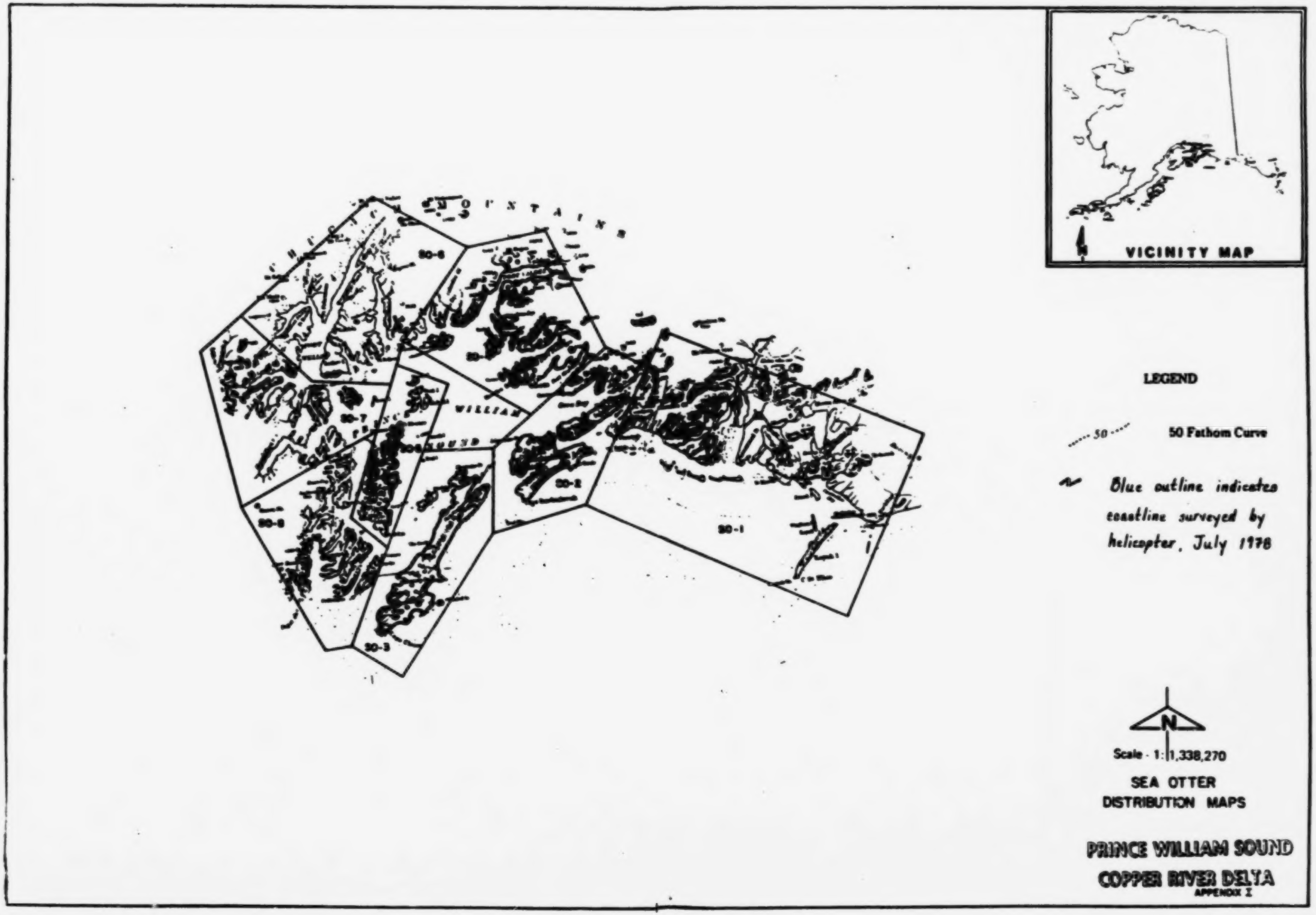
MOUNTAINS
WILLIAM
SOUND
30-1
30-2
30-3
30-6
30-7
30-8
VICINITY MAP
LEGEND
50
50 Fathom Curve
Blue outline indicates coastline surveyed by helicopter, July 1978
N
Scale - 1:1,338,270
SEA OTTER DISTRIBUTION MAPS
PRINCE WILLIAM SOUND
COPPER RIVER DELTA
APPENDIX I

Figure 5. Map of Prince William Sound showing an overview of the area surveyed for sea otters. The heavy line indicates area surveyed.

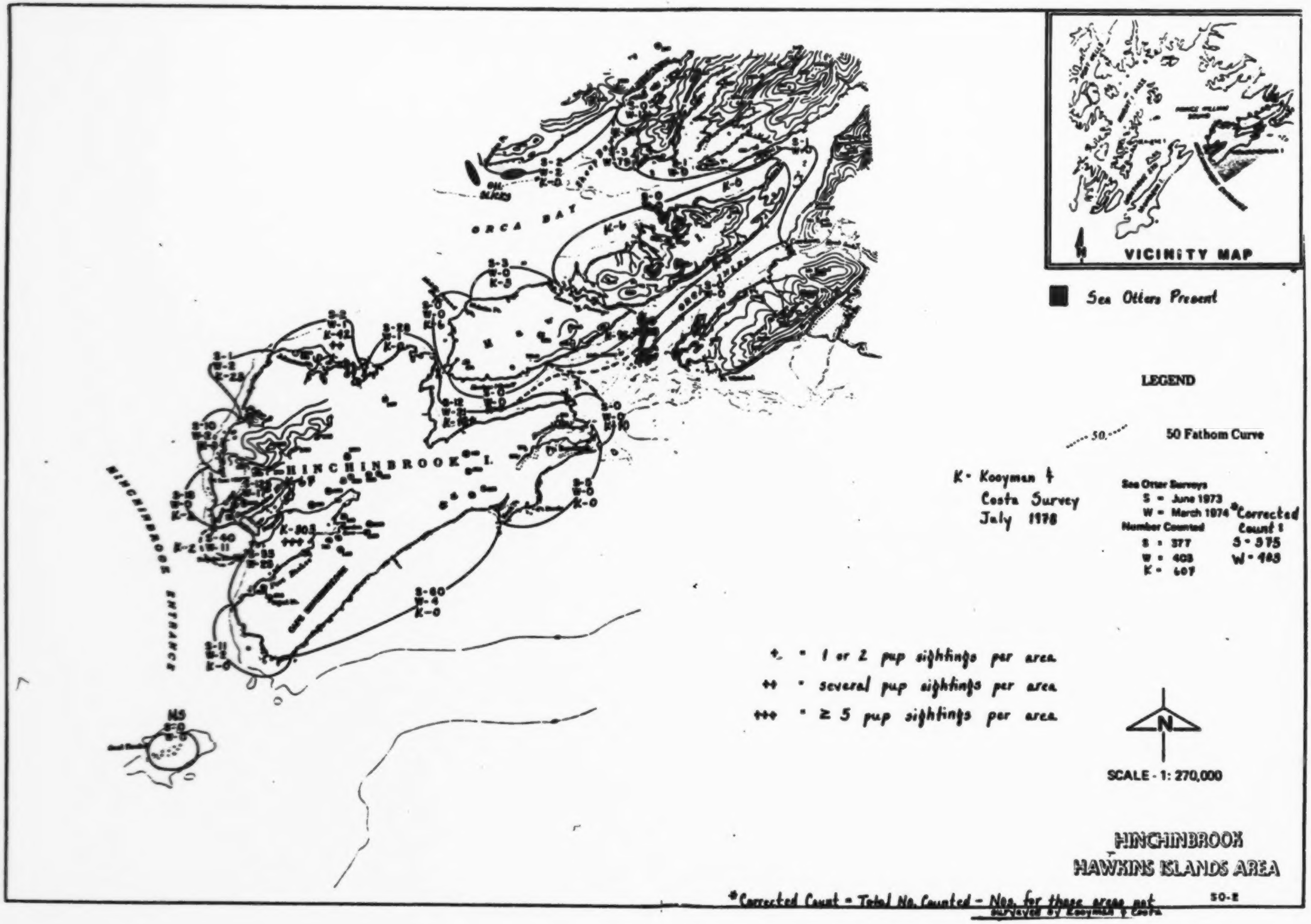
VICINITY MAP
Sea Otters Present
LEGEND
50
50 Fathom Curve
K - Kooyman & Costa Survey July 1978
Sea Otter Surveys
S = June 1973
W = March 1974
Number Counted
S = 377
W = 403
K = 607
*Corrected Count !
S = 375
W = 405
+ = 1 or 2 pup sightings per area
++ = several pup sightings per area
+++ = ≥ 5 pup sightings per area
N
SCALE - 1: 270,000
HINCHINBROOK
HAWKINS ISLANDS AREA
*Corrected Count = Total No. Counted - Nos. for those areas not surveyed by Kooyman & Costa
50-2
ORCA BAY
OIL SLICKS
HINCHINBROOK I.
HINCHINBROOK ENTRANCE
K-6
K-0
K-303
+++
K-2
S-40
W-11
S-60
W-4
K-0
S-11
W-2
K-0
S-5
W-0
K-0
S-3
W-0
K-3
S-1
W-2
K-23

Figure 6. Displays sea otter sightings for Hinchinbrook and Hawkins Islands area. S and W are sightings reported by Calkins, Pitcher and Schneider (1975).

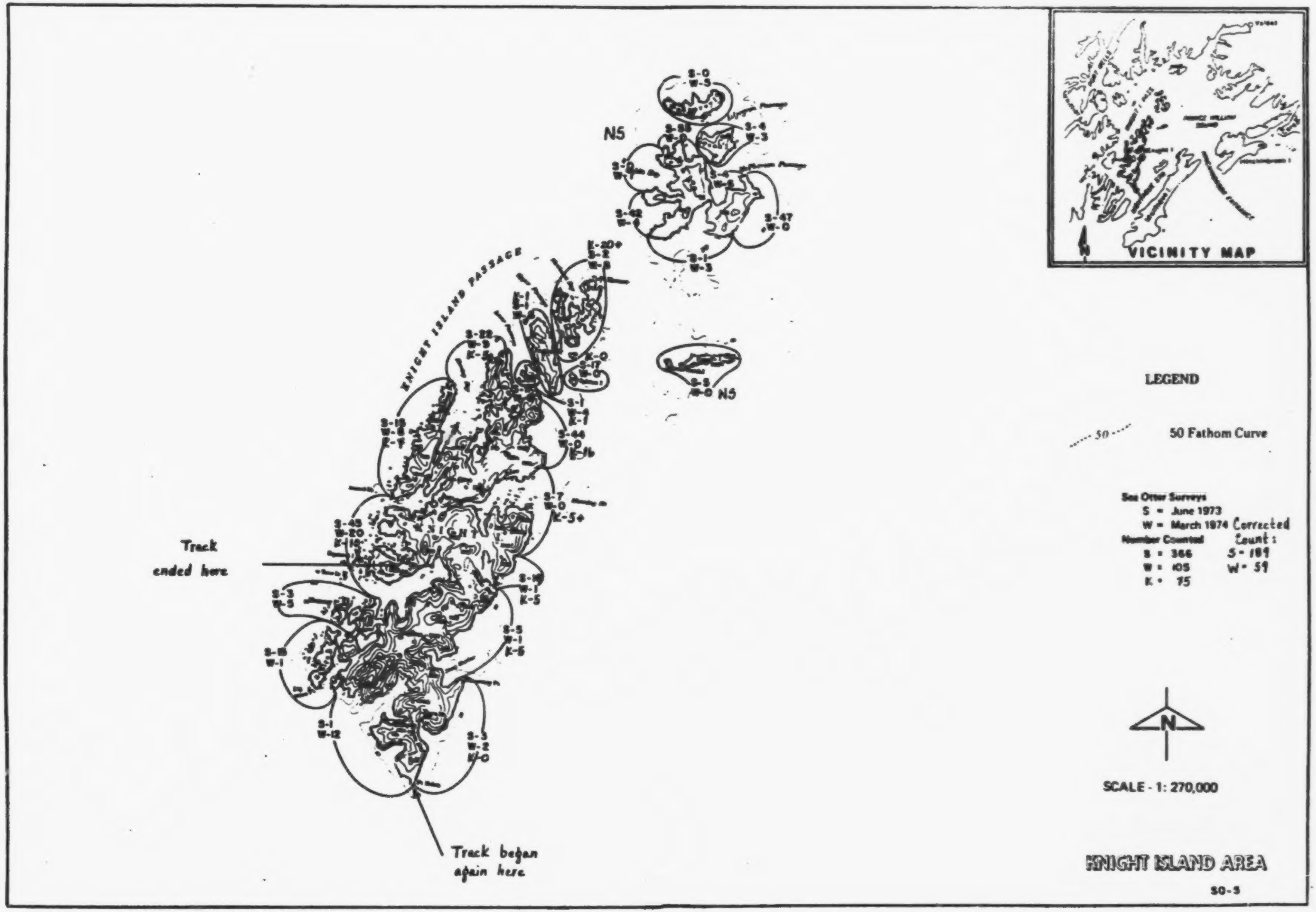
VICINITY MAP
N
S-0
W-3
NS
S-4
W-3
S-42
W-6
S-47
W-0
S-1
W-3
K-20+
S-2
W-8
KNIGHT ISLAND PASSAGE
S-22
W-9
K-5
K-0
S-17
W-0
S-5
W-0
NS
S-1
W-7
K-7
S-18
W-6
K-7
S-44
W-0
K-16
S-7
W-0
K-5+
S-45
W-20
K-10
KNIGHT
Track
ended here
S-3
W-5
S-18
W-1
K-5
S-5
W-1
K-6
S-15
W-1
S-1
W-12
S-3
W-2
K-0
Track began
again here
LEGEND
50
50 Fathom Curve
Sea Otter Surveys
S = June 1973
W = March 1974
Number Counted
S = 366
W = 105
K = 75
Corrected Count:
S - 189
W - 59
N
SCALE - 1: 270,000
KNIGHT ISLAND AREA
SO-3

Figure 7. Displays sea otter sightings for Knight Island area. S and W are sightings reported by Calkins, Pitcher and Schneider (1975)

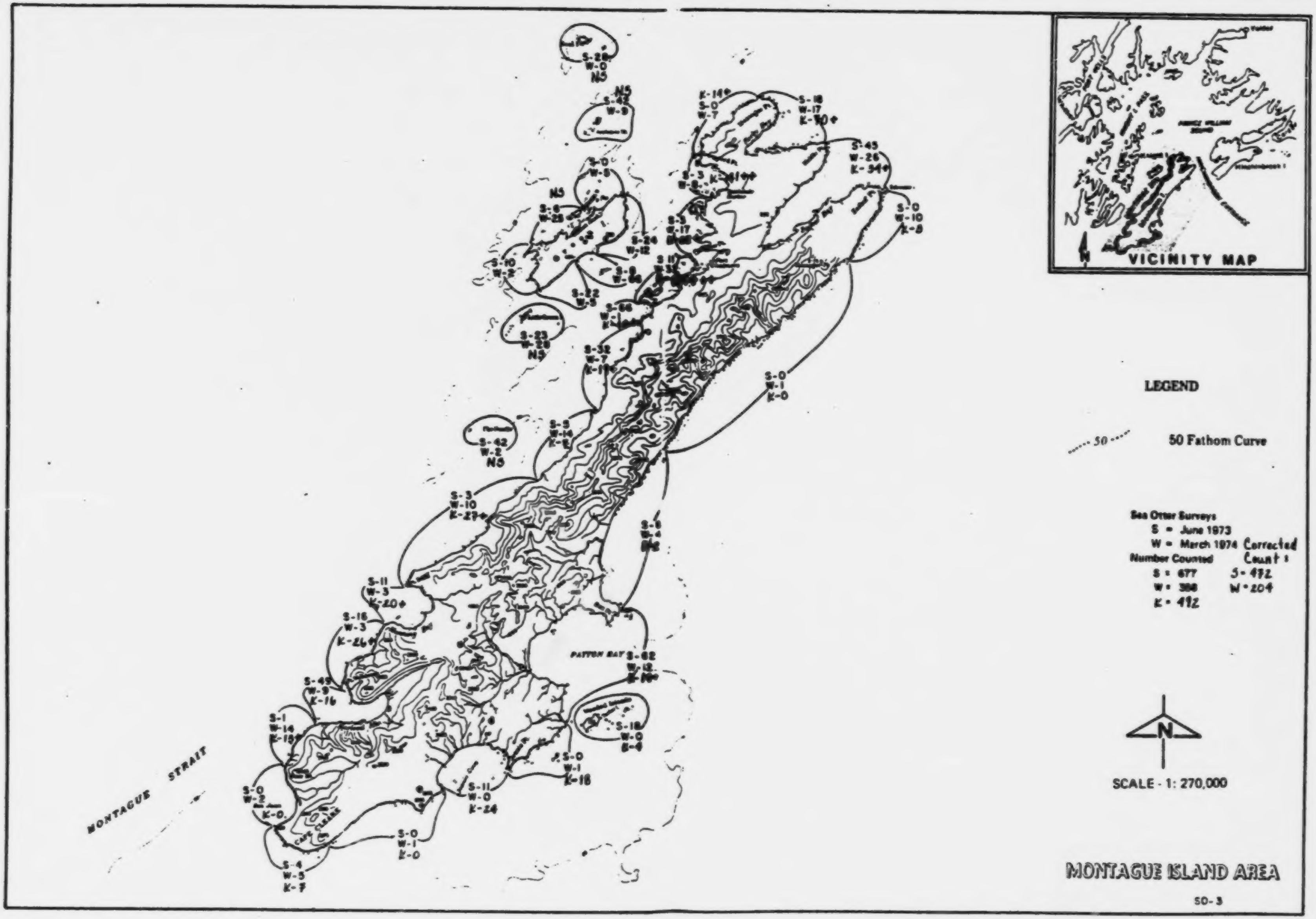
VICINITY MAP
S-28
W-0
NS
NS
S-42
W-9
K-14+
S-0
W-7
S-18
W-17
K-50+
S-45
W-26
K-34+
S-0
W-5
NS
S-6
W-25
S-3
W-8
K-61++
S-0
W-10
K-8
S-10
W-2
S-24
W-12
S-5
W-17
S-22
W-5
S-23
W-28
NS
S-32
W-7
K-11+
S-0
W-1
K-0
S-42
W-2
NS
S-5
W-14
K-8
S-3
W-10
K-27+
S-11
W-3
K-20+
S-16
W-3
K-26+
S-49
W-9
K-16
S-1
W-14
K-15+
S-0
W-2
K-0
S-4
W-5
K-7
S-0
W-1
K-0
S-11
W-0
K-24
S-0
W-1
K-18
S-18
W-0
PATTON BAY
S-82
W-12
CAPE CLEARE
MONTAGUE STRAIT
LEGEND
50
50 Fathom Curve
Sea Otter Surveys
S = June 1973
W = March 1974
Number Counted
S = 677
W = 388
K = 492
Corrected Count:
S = 492
W = 204
N
SCALE - 1: 270,000
MONTAGUE ISLAND AREA
SO-3

Figure 8. Displays sea otter sightings for Montague Island area. S & W are sightings reported by Calkins, Pitcher and Schneider (1975)

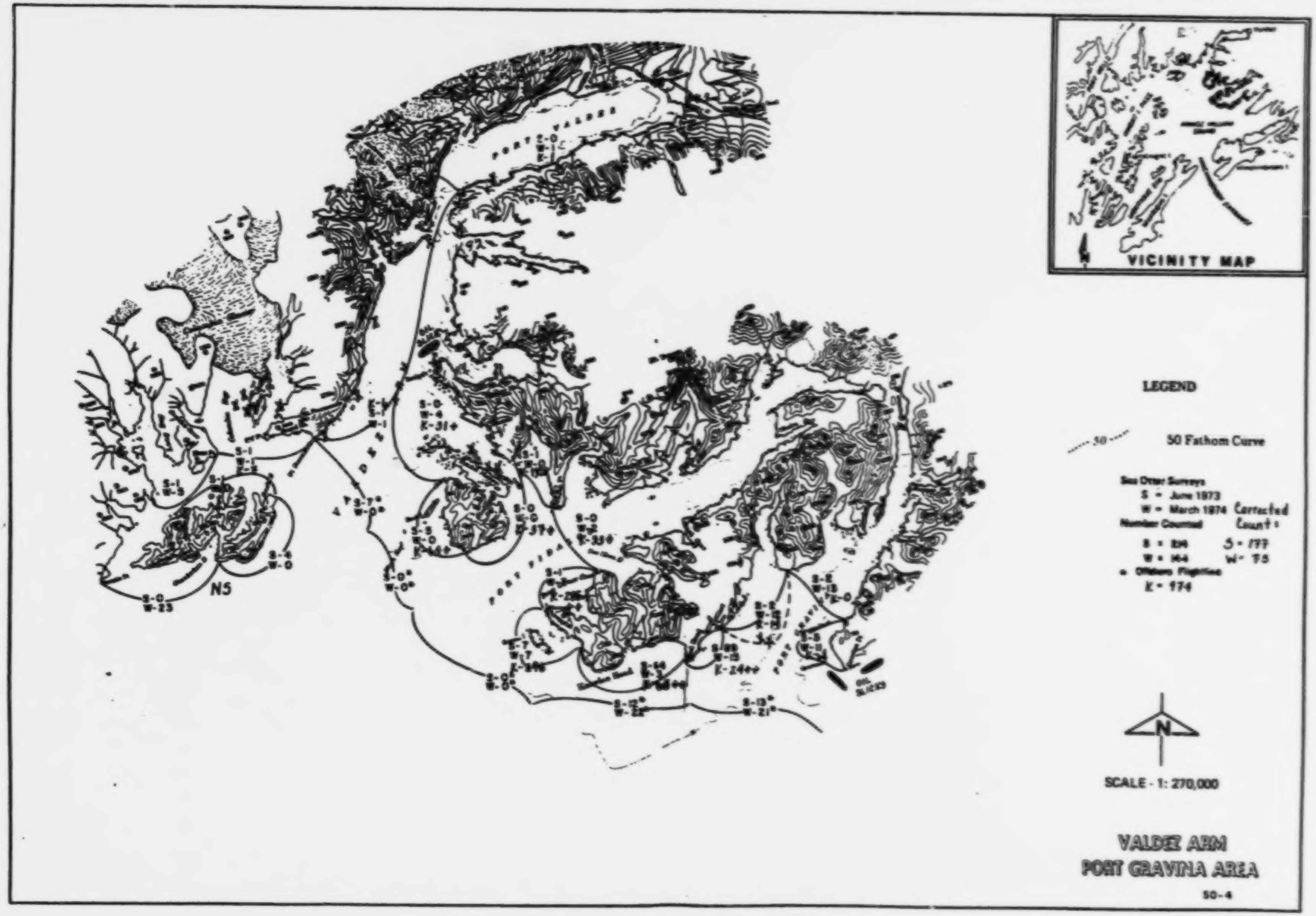
PORT VALDEZ
S-0
W-1
NS
S-0
W-23
VICINITY MAP
LEGEND
50 Fathom Curve
Sea Otter Surveys
S = June 1973
W = March 1974
Number Counted
S = 234
W = 144
Corrected Count:
S = 177
W = 75
• Offshore Flightline
K = 174
N
SCALE - 1: 270,000
VALDEZ ARM
PORT GRAVINA AREA
50-4

Figure 9. Displays sea otter sightings for Valdez Arm-Point Gravina area. S and W are surveys conducted by Calkins, Pitcher and Schneider (1975)

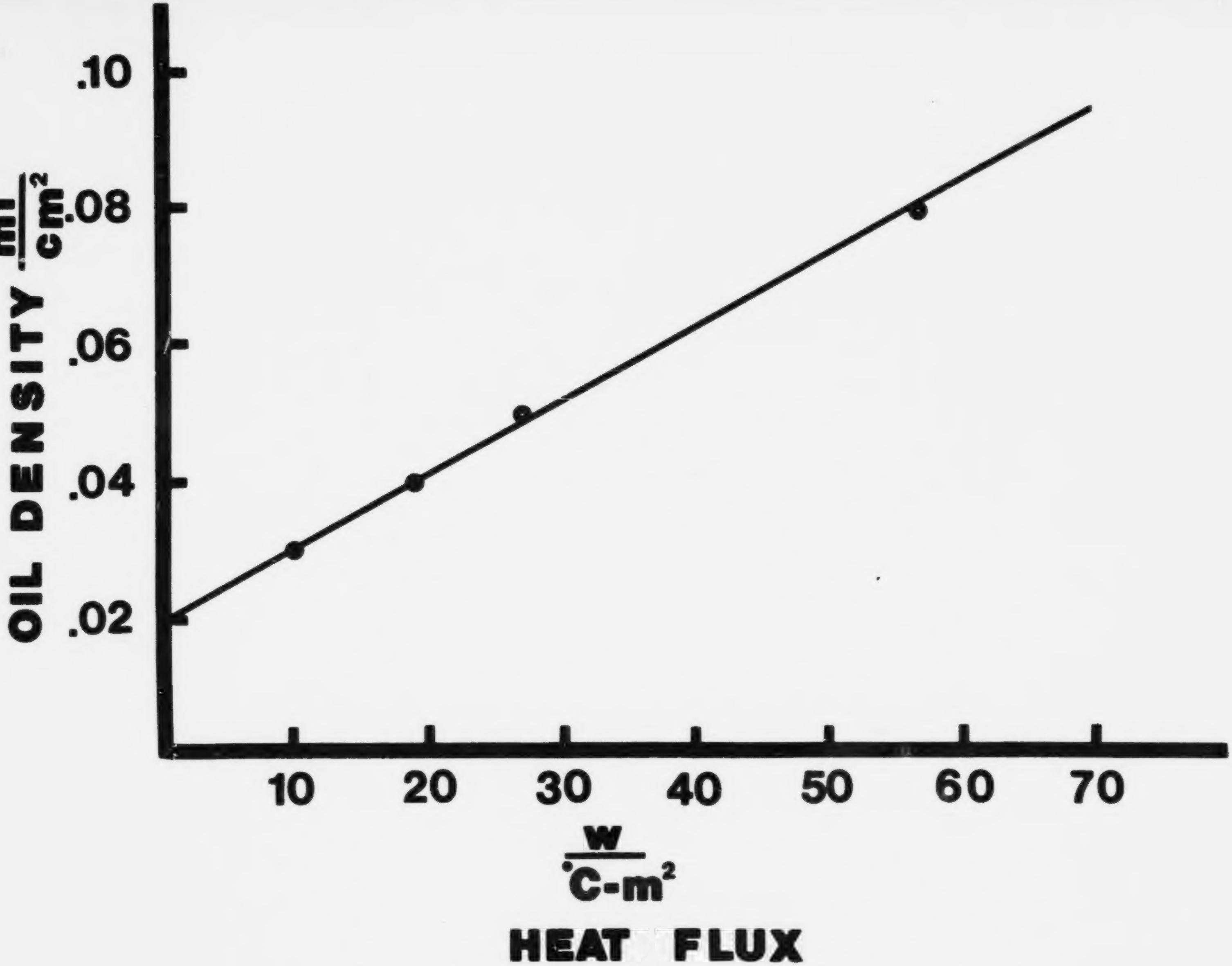

Figure 10. The relative amount of oil applied in ml per unit area of fur covered in cm^2 (oil density ml/cm^2) is plotted against the increased heat flux through the oiled fur calculated for the four oiling experiments.

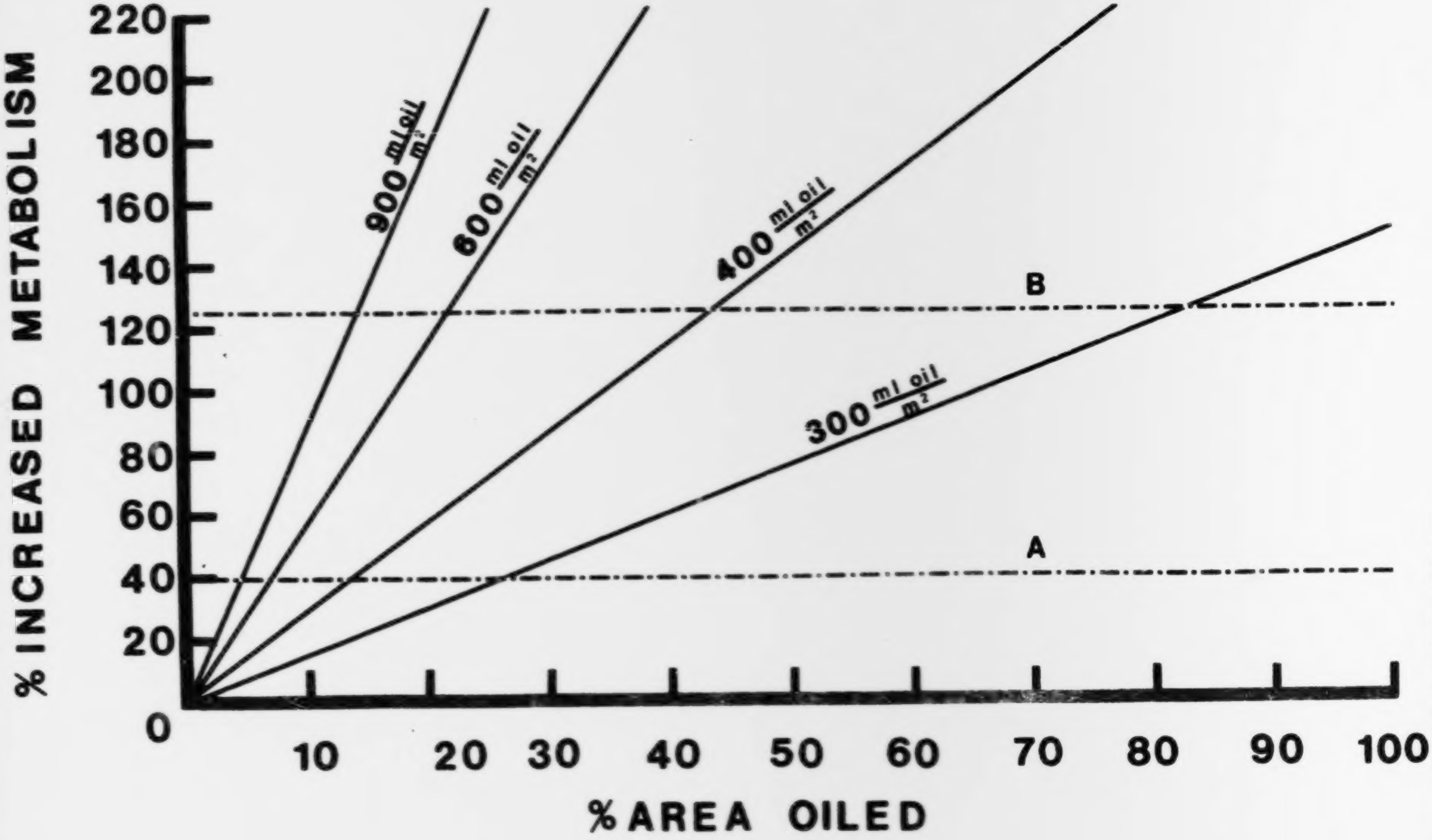

Figure 11. The predicted increases in resting metabolism are plotted against the percent of the sea otter body surface oiled for 3 densities of oil contamination. For comparison, the highest increase in resting oxygen consumption after oiling (otter 1, line A) and the highest resting oxygen consumption recorded for any condition (otter 5, washed , line B) are presented.

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Final Report

Reconnaissance of Intertidal Communities in the Eastern Bering Sea and the Effects of Ice-scour on Community Structure

by

Charles E. O'Clair, Joyce L. Hanson, Richard T. Myren,
Jessica A. Gharrett, Theodore R. Merrell, Jr., and
John S. MacKinnon with Appendix on

Some Benthic Marine Algae from the Pribilof Islands, Bering Sea: A Preliminary Annotated List

by

Natasha I. Calvin

Northwest and Alaska Fisheries Center Auke Bay Laboratory
Outer Continental Shelf Energy Assessment Program

Sponsored by

U.S. Department of the Interior
Bureau of Land Management

1 October 1979

Preface

At least one of us was present and participated in collecting data at each site. When possible, the description of the intertidal community at a particular site was written by an individual who had participated in sampling that site because firsthand familiarity with the physical setting, sampling methods, and the general patterns of distribution of major organisms contributes to a fuller interpretation of the data from quantitative sampling. The senior author did not participate in any of the field work so those descriptions written by him have not benefited from personal observations in the field. The name of each locality sampled is listed in the Table of Contents and is followed by the initials of the author(s) who described the biotic communities at that locality.

Table of Contents

Page
Preface 110

List of Figures 113

List of Tables 117

Acknowledgements 120

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development 121

II. Introduction 123

- A. General nature and scope of study 123
- B. Specific objectives 124
- C. Relevance to problems of petroleum development 125

III. Current state of knowledge 127

IV. Study areas 129

V. Sources, methods, and rationale of data collection 130

- A. Field methods 130
- B. Laboratory procedures 133
- C. Sources of bias and error 134

VI. Results 135

- A. Distributions and relative abundances of intertidal biota 135
  - 1. Bristol Bay and eastern Aleutian Islands 135
    - 1.1 Nushak Island (JH) 135
    - 1.2 Spectacle Island (JH) 142
    - 1.3 Middle Point (JH) 145
    - 1.4 Point Edward (JH) 147
    - 1.5 Moffet Lagoon (RM/JG) 150
    - 1.6 Blaine Point (CO/JG) 150
    - 1.7 Izembek Lagoon (RM) 154
    - 1.8 Operl Island (TM) and Cape Glazenap (RM/JG) 160
    - 1.9 Amak Island (JH) 164
    - 1.10 Cape Lapin (RM) 167
    - 1.11 Cape Mordvinof (JH/JG) 175
    - 1.12 Sennet Point (CO/JG) 179
    - 1.13 Akun Island (JH) 184
    - 1.14 Eider Point (RM) 187
    - 1.15 Portage Bay (JH) 197
    - 1.16 Crooked Island (CO/JG) 203
    - 1.17 Cape Peirce (CO/JG) 207

2. Pribilof Islands ---------- 214
    2.1 St. Paul Island ---------- 214
        2.1.1 English Bay (TM) ---------- 214
        2.1.2 North Beach and East Beach (TM) ---------- 218
    2.2 Otter Island (JM) ---------- 219
    2.3 St. George Island ---------- 222
        2.3.1 Garden Cove (RM) ---------- 222
        2.3.2 High Bluffs (RM) ---------- 227
        2.3.3 Zapadni Bay (JM) ---------- 228
3. Norton Sound ---------- 232
    3.1 Stuart Island (CO/JG) ---------- 236
    3.2 Wood Point (CO/JG) ---------- 241
    3.3 Malikfik Bay (JH) ---------- 246
    3.4 Cape Denbigh (CO/JG) ---------- 252
    3.5 Golovnin Lagoon (CO/JG) ---------- 255
    3.6 Rocky Point (CO/JG) ---------- 259
    3.7 Square Rock (CO/JG) ---------- 262
    3.8 Bluff (CO/JG) ---------- 262
    3.9 Safety Sound (CO/JG) ---------- 267
    3.10 Cape Nome (CO/JG) ---------- 267
    3.11 Sledge Island (JH) ---------- 272
    3.12 Cape Woolley (CO/JG) ---------- 277
    3.13 King Island (JH) ---------- 283

B. Effects of scouring by sea ice on intertidal community structure ---------- 287

1. Physical disturbance by sea ice ---------- 287
    1.1 Island biogeography ---------- 290
    1.2 Species richness ---------- 293
    1.3 Species-area curves ---------- 296
    1.4 Species-importance curves ---------- 298

2. Effects on community dominants and succession ---------- 303
3. Competition, herbivory, and community development after ice-scouring ---------- 310
4. Populations of sessile invertebrates ---------- 319
5. Refuges from ice-scouring ---------- 322

VII. Discussion ---------- 326

VIII. Conclusions ---------- 329

IX. Needs for further study ---------- 330

X. References ---------- 331

XI. Appendix I ---------- 340
    Appendix II ---------- 342
    Appendix III ---------- 393

Errata ---------- 414

List of Figures

		Page
Fig. 1.	Location of sites in the western Gulf of Alaska, Bristol Bay, and on the Aleutian Islands.	136
Fig. 2.	Location of site at Nukshak Island.*	138
Fig. 3.	Nukshak Island sampling area. Site of Transect 1.	140
Fig. 4.	Location of site at Spectacle Island.*	143
Fig. 5.	Replicate transect lines at Spectacle Island site.	144
Fig. 6.	Arrow sampling site at Spectacle Island.	144
Fig. 7.	Location of sites at Point Edward and Middle Point.*	146
Fig. 8.	Core sampling at Point Edward.	148
Fig. 9.	Location of sites at Operl Island, Izembek Lagoon, Blaine Point, and Moffet Lagoon.*	151
Fig. 10.	Biomass in core samples taken at Moffet Lagoon.	152
Fig. 11.	Blaine Point site.	153
Fig. 12.	Elevation, number of species, and eelgrass biomass (wet weight) for the transect stations at Izembek Lagoon.	157
Fig. 13.	Comparative biomass of dominant pelecypods and annelids found in transect stations at Izembek Lagoon.	158
Fig. 14.	Operl Island sand sample site.	162
Fig. 15.	Operl Island site showing magnetic sand, razor clam shells, and pumice rocks. Light-colored patch about 2 m in diameter to left of figure is black magnetic sand.	162
Fig. 16.	Location of sites at Cape Glazenap.*	163
Fig. 17.	Location of site at Amak Island.*	165
Fig. 18.	View of transect at Amak Island sampling site.	166
Fig. 19.	Location of site at Cape Lapin.*	168
Fig. 20.	Mean biomass per 1/16 m^2 quadrat in randomized meters within lateral transects at Cape Lapin.	173
Fig. 21.	Location of the site at Cape Mordvinof.*	176
Fig. 22.	Cape Mordvinof sampling site.	178

* These figures modified from Sears and Zimmerman (1977).

BEST DOCUMENT AVAILABLE

Fig. 23. Location of site at Sennet Point.* 180

Fig. 24. Site at Sennet Point; view of upper portion of Transect 2. 182

Fig. 25. Location of site at Akun Island.* 185

Fig. 26. Akun Island, Aleutian Islands. View of sampling area at low tide. 186

Fig. 27. Location of site at Eider Point.* 188

Fig. 28. Site at Eider Point. 189

Fig. 29. View of the *Alaria* stratum and Transect 4 at Eider Point. 189

Fig. 30. Biomass per 1/16 m^2 quadrat in systematic samples (Transect 1) at Eider Point. 191

Fig. 31. Mean biomass per 1/16 m^2 quadrat in random m^2 plots along Transect 4 (the *Alaria* stratum) at Eider Point. 195

Fig. 32. Location of site at Portage Bay.* 198

Fig. 33. Aerial view of sampling site at Portage Bay. 199

Fig. 34. Portage Bay sampling area. 199

Fig. 35. Typical invertebrate assemblage in *Balanus cariosus* zone at Portage Bay site. 200

Fig. 36. View of sampling area at Portage Bay showing part of transect line, zonation study site, and plethora of workers. 200

Fig. 37. Concentration of littorine snails along supralittoral fringe at Portage Bay sampling site. 204

Fig. 38. Location of site at Crooked Island.* 205

Fig. 39. Looking seaward along the transect line at Crooked Island. 206

Fig. 40. Location of site at Cape Peirce.* 209

Fig. 41. View of the bedrock mound site at Cape Peirce. 210

Fig. 42. View of zonation study (arrow) site at Cape Peirce. 210

Fig. 43. Location of study sites in the Pribilof Islands. 215

Fig. 44. Location of the three St. Paul Island sites: English Bay, North Beach, and East Beach.* 217

Fig. 45. Location of the study site at Otter Island.* 220

* These figures modified from Sears and Zimmerman (1977)

Fig. 46. Upper half of Transect 1-75, Otter Island. 221

Fig. 47. Lower end of Transect 1-75 at Otter Island. 223

Fig. 48. Lower end of Transect 1-76 at Otter Island. 223

Fig. 49. Location of the three St. George Island sites: Zapadni Bay, High Bluffs, and Garden Cove.* 224

Fig. 50. Study sites at Garden Cove. 225

Fig. 51. Transect sites at Garden Cove. 226

Fig. 52. Lower end of Transect 1 at Garden Cove. 226

Fig. 53. Upper end of systematic transect at Zapadni Bay. 229

Fig. 54. Lower half of systematic transect at Zapadni Bay. 229

Fig. 55. Location of sites in Norton Sound. 233

Fig. 56. Location of the site at Stuart Island.* 238

Fig. 57. The transect at Stuart Island. 239

Fig. 58. Location of the study area at Wood Point.* 243

Fig. 59. Lower end of transect at Wood Point. 244

Fig. 60. Upper end of transect at Wood Point. 245

Fig. 61. Location of the Cape Denbigh and Malikfik Bay study sites.* 249

Fig. 62. Wetlands in the vicinity of Malikfik Bay. 250

Fig. 63. Study area on a beach seaward of Malikfik Bay. 250

Fig. 64. Bivalve shells in drift at Malikfik Bay. 253

Fig. 65. Location of sites at Rocky Point and Golovnin Lagoon.* 256

Fig. 66. Study site at Golovnin Lagoon. 257

Fig. 67. Study site near Rocky Point. 260

Fig. 68. Location of sites at Bluff Point and Square Rock.* 263

Fig. 69. View to the east from the study area at Square Rock. 264

Fig. 70. View east from the study site at Bluff. 266

Fig. 71. Location of sites at Cape Nome and Safety Sound.* 269

* These figures modified from Sears and Zimmerman (1977).

Fig. 72. Study site near the entrance of Safety Sound. 270

Fig. 73. Location of the site at Sledge Island.* 274

Fig. 74. Sledge Island study site, Norton Sound. 275

Fig. 75. Fucus sp. growing between boulders below MLLW at Sledge Island. 278

Fig. 76. Littorina sp. grazing algae on boulder at Sledge Island. 278

Fig. 77. Location of site at Cape Woolley.* 280

Fig. 78. Study site on a sand beach north of Cape Woolley. 281

Fig. 79. Rock at Cape Woolley covered with Fucus, Chthamalus, and an unidentified filimentous alga. 281

Fig. 80. Location of the study site at King Island. 284

Fig. 81. Boulder-strewn intertidal area at King Island. 285

Fig. 82. Intertidal sampling area below abandoned village at King Island. 286

Fig. 83. Species-area curves of mollusks in quadrats collected in the intertidal region at four islands in the Bering Sea. 297

Fig. 84. Relationship between relative importance and rank of mollusks at four islands, the shores of two of which are frequently scoured by sea ice. 301

Fig. 85. Mean weights of four species of algae at four islands in the Bering Sea. 311

Fig. 86. Mean weights of seven species of benthic herbivore at ice-scoured and unscoured islands in the Bering Sea. 313

Fig. 87. Mean weight of sessile invertebrates at ice-scoured and unscoured islands in the Bering Sea. 320

Fig. 88. Intertidal region at English Bay. 323

Fig. 89. Garden Cove site showing the deep fissures in and crevices between rocks. 324

* These figures modified from Sears and Zimmerman (1977).

List of Tables

Page

Table 1. Pertinent sampling information for 18 sites in the western Gulf of Alaska, Bristol Bay, and the Aleutian Islands. 137

Table 2. Frequency of occurrence of selected species along four transect lines on Nukshak Island. 141

Table 3. Statistics for the ten species of invertebrates showing the greatest average rank on the transect at Point Edward. 149

Table 4. Statistics for the ten species of plants and invertebrates showing the greatest average rank at the transect at Blaine Point. 155

Table 5. Analysis of variance of linear regression for number of species vs. wet weight of eelgrass at Izembek Lagoon. 161

Table 6. Total biomass and number of species or groups in pairs of 1/16 m^2 quadrats taken from m^2-plots in three strata at Cape Lapin. 170

Table 7. Key to the species and groups identified in Cape Lapin samples. 171

Table 8. Numbers of individuals in the dominant and secondary biomass categories by stratum, Cape Lapin. 174

Table 9. Statistics for the ten species of plants and invertebrates showing the greatest average rank at the transect at Sennet Point. 183

Table 10. Constancy, dominance, and tidal range of three species of barnacles collected at Portage Bay, Alaska. 202

Table 11. Statistics for the 11 species of algae and invertebrates showing the greatest average rank on the transect at Crooked Island. 208

Table 12. Statistics for the ten species of algae and invertebrates showing the greatest average rank in quadrats at Cape Peirce. 212

Table 13. Pertinent sampling information for seven sites in the Pribilof Islands. 216

Table 14. Geographic coordinates, sampling dates, and beach and surface seawater characteristics at 13 localities in Norton Sound where intertidal communities were sampled. 234

Table 15. Pertinent sampling information for 14 intertidal sites in Norton Sound. 235

Table 16. Average species counts of plants and invertebrates in quadrats or cores at intertidal sites in Norton Sound. 237

Table 17. Abundances of species of algae and invertebrates on the transect at Stuart Island. 240

Table 18. Statistics for ten species of algae and invertebrates which had the greatest average rank in quadrats at Stuart Island. 242

Table 19. Abundances of the most frequently occurring species of algae and invertebrates on the rocky intertidal transect at Wood Point. 247

Table 20. Abundances of species of algae and invertebrates at the peat site at Wood Point. 248

Table 21. Abundances of species of algae and invertebrates in 1-liter cores at Malikfik Bay. 251

Table 22. Abundances of invertebrates in sand samples from Cape Denbigh. 254

Table 23. Abundances of plants and invertebrates in eight 7-liter samples from a sand beach at Golovnin Lagoon. 258

Table 24. Abundances of plants and invertebrates in five 8-liter samples from a sand beach at Rocky Point. 261

Table 25. Abundances of invertebrates in three 8-liter samples from a gravel beach at Square Rock. 265

Table 26. Abundances of plants and invertebrates in six 7-liter samples from a gravel and sand beach at Bluff. 268

Table 27. Abundances of plants and invertebrates in six 8-liter samples from a sand and mud beach at Safety Sound. 271

Table 28. Abundances of plants and invertebrates in two 7-liter samples from a sand beach near Cape Nome. 273

Table 29. Abundances of species of algae and invertebrates in samples from a sand beach at Sledge Island. 276

Table 30. Abundances of plants and invertebrates in four 7-liter samples from a sand beach at Cape Woolley. 282

Table 31. Sea ice at the Pribilof Islands and Amak Island in five recent winters. 288

Table 32. Average species counts of plants and invertebrates in quadrats at rocky intertidal areas in the Pribilof Islands and at Amak and Akun Islands. 294

Table 33. Analysis of variance of species counts of algae and invertebrates at upper and lower rocky intertidal levels in the Pribilof Islands and at Amak and Akun Islands. 294

Table 34. Three orthogonal comparisons of species counts of algae and invertebrates in the rocky intertidal region at the Pribilof Islands and at Amak and Akun Islands. 295

Table 35. Smirnov test of differences in species-area curves for mollusks in the intertidal region at St. George, Otter, Amak, and Akun Islands. 299

Table 36. Dominant (in biomass) species, frequency of dominance, and frequency of occurrence of each species in upper and lower intertidal zones at Amak, Akun, and the Pribilof Islands. 306

Table 37. Ephemeral species and mean wet weight in upper and lower intertidal zones at Amak, Akun, and the Pribilof Islands. 307

Table 38. Canopy species and mean wet weight in upper and lower intertidal zones at Amak, Akun, and the Pribilof Islands. 308

Table 39. Food of *Littorina*. 314

Table 40. Tests of significance of *Littorina* wet weights (*L. sitkana* and *L. aleutica* combined) in the rocky intertidal region at the Pribilof Islands compared with Amak and Akun Islands. 317

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I. SUMMARY OF OBJECTIVES, CONCLUSIONS, AND IMPLICATIONS WITH RESPECT TO OCS OIL AND GAS DEVELOPMENT.

The intertidal region of most shores in the eastern Bering Sea north of 56°N is subject to scouring by sea ice in late winter and spring in most years. We used data collected with systematically sampled belt transects and arrays of randomly placed quadrats to compare intertidal communities on rocky shores in the Pribilofs with those on the shores of islands in the southeastern Bering Sea that have not been subject to recent scouring. We used the above methods and randomly sampled belt transects and haphazard sampling to study patterns of distribution and abundance of macrobiota at 39 intertidal sites in the eastern Bering Sea.

In mid-summer 1975, 4 months after the most recent scouring episode in the Pribilof Islands, species richness of most major taxa of plants and invertebrates generally and Mollusca specifically (as determined by average species counts and species-area curves respectively) was significantly lower in the Pribilof Islands than at Amak and Akun Islands (the shores of which had not been recently scoured by ice). Curves of the distribution of biomass among species of Mollusca were markedly different at the Pribilof Islands as compared with those for Amak and Akun Islands. The Pribilof Island curves showed a greater concentration of dominance among a few species.

Fugitive species of algae had the greatest wet weight in most quadrats at the Pribilof Islands whereas canopy species were preponderant on unscoured shores. The biomass of ephemeral algae and of known consumers of ephemeral algae was about the same at all sites. Sessile invertebrates were usually small and low in abundance on, or absent from, unprotected surfaces at the Pribilof Islands.

The availability of refuges from ice-scouring probably allows many species which otherwise would be eliminated to survive in the intertidal community at the Pribilof Islands. The chief impact of oil pollution on the community would most likely be to reduce or deny access to these refuges by intertidal organisms. All of the three types of refuges from ice-scouring (temporal periods outside the activity range of ice-scouring, spatial zones beyond the activity range of ice-scouring, and physical heterogeneities within the activity range of ice-scouring) available to intertidal organisms could be adversely affected by an oil spill. The extent to which the refuges would be denied to the organisms would depend on the timing of the oil spill and to a lesser extent on the viscosity of the oil.

II. INTRODUCTION

A. General nature and scope of study.

The primary goal of our studies has been to generally characterize intertidal communities in the Gulf of Alaska and eastern Bering Sea. The studies were designed to provide baseline information on the patterns of distribution and relative abundance of plants and invertebrates in these communities prior to the development of oil and gas leases on the continental shelf in these regions.

B. Specific objectives.

Our specific objectives have been to determine the composition of intertidal communities and to describe the distributions and abundances of species in them at as many representative sites as possible near the proposed oil and gas lease areas. In addition we have examined possible mechanisms of community organization at our sites and how they might determine the response of a community to oil spills.

In this report, we describe the intertidal communities at our study sites in the Bering Sea (those in the Gulf of Alaska have been described elsewhere [O'Clair et al. 1978 and Zimmerman et al. 1978]), and examine the role of scouring by sea ice in structuring intertidal communities in the Bering Sea.

C. Relevance to problems of petroleum development.

Our descriptions of intertidal communities in the Gulf of Alaska and the Bering Sea should provide information on the character of natural, unpolluted communities at our study sites. Our results can be compared at some future date with measurements of oiled communities at the same sites should an oil spill occur. We can be reasonably confident that if a spill is sufficiently devastating to produce profound changes in the species composition at our study sites we should be able to document those changes with the information at hand. However, our confidence plummets when we consider subtler differences such as changes in the abundance or biomass of particular species or if we attempt to extrapolate our results to nearby localities or different seasons. Our lack of confidence derives from an appreciation of the enormous temporal and spatial variability in biological systems in nature. Mann and Clark (1978) recently reiterated this problem with respect to documenting the effects of oil spills on marine intertidal communities. Not surprisingly, Zimmerman et al. (1978) found that Alaskan intertidal communities are also highly variable. Some of this variation is stochastic and therefore unpredictable, but as Pielou (1972) points out, if we avoid errors resulting from (1) a dearth of factual knowledge and (2) faulty reasoning, then we can be fairly successful in predicting ecological change in natural systems. Until we can do this we have no hope of adequately assessing the effects of oil spills on biotic communities.

The most fruitful way of approaching the predictable domain of community dynamics is through the study of the most important mechanisms controlling community organization. These usually include competition, predation, and physical disturbance. In a previous report (O'Clair et al. 1978) we briefly reviewed evidence suggesting the importance of understanding key interspecific interactions to the assessment of the impact of oil pollution on marine communities and considered how dominant competitors might influence the number and relative abundance of other species in the community. Here we concentrate on the role of physical disturbance, specifically scouring by sea ice.

Scouring by sea ice is an important physical disturbance on most shores in the eastern Bering Sea north of 56°N. It is an annual occurrence which can scrape most organisms from upper surfaces on rocky shores and can plow up organisms in unconsolidated sediments. Systems affected by ice scouring are generally species-poor (McRoy and Allen 1974). Presumably, most of those species which persist in the system have evolved life history strategies which allow them to either escape the effects of ice-scouring or to recolonize scoured surfaces rapidly from refuges (such as crevices and spaces beneath and between boulders) in the intertidal or subtidal regions. Oil pollution might be expected to further stress those organisms that have evolved strategies to cope with ice scouring and thereby further reduce species richness in an already simple system. Moreover, because it can penetrate crevices and the interstices of boulder fields, oil could have a far greater impact on the community by making refuges uninhabitable to both sessile and motile species. Before we can properly evaluate the effects of oil on ice-scoured systems we should determine such things as the effects of scouring on community composition and trophic structure, the rates and trajectories of ecological succession following a scouring episode and the roles played by competition and predation in determining these rates and trajectories. Once we understand the properties of unpolluted ice-scoured systems, we can then use our knowledge of relative susceptabilities of organisms in the system to oil toxicity, rates of decomposition of oil in cold climates, and other factors to evaluate the effect of an oil spill on the system.

III. CURRENT STATE OF KNOWLEDGE

Local patterns of species diversity are frequently controlled by disturbance. As used here, a disturbance includes any event that sets back the progress of ecological succession to an earlier stage. Levin and Paine (1974) have developed a model which predicts the species richness of a community from information on the size, age, and distribution of patches of open space created by disturbance. Two general predictions of their model are that disturbances that are localized in space and time should increase species richness whereas disturbances that are frequent, chronic, or severe should simplify community structure and reduce species richness. These predictions are supported by empirical studies in marine (Dayton 1971) and terrestrial (Loucks 1970, Taylor 1973) systems.

Connell (1978) has proposed a conceptual model that accounts for the high diversity of trees in tropical rain forests and corals on tropical reefs by an intermediate disturbance mechanism. Under this model, disturbances of intermediate frequency and magnitude promote high diversity, whereas high or low frequencies or magnitudes of disturbance result in lower diversity either by preventing late-arriving and slow-growing species from colonizing or by allowing the community to develop toward a low-diversity equilibrium state.

A general hypothesis of species diversity has been offered by Huston (1979) who claims that knowledge of the population growth rates of competitors and the frequency of population reductions caused by predation or disturbance is necessary to predict the diversity of a community. In all of these models, disturbance promotes high species diversity by preventing the system from reaching competitive equilibrium.

McRoy and Allen (1974) review the available literature on ice-stressed ecosystems. We examine the effects of ice scour on rocky intertidal communities because our observations in the Bering Sea indicate that these communities tend to be more diverse than those on sandy or gravelly beaches, and we would expect a diverse community to be a more sensitive indicator of the magnitude of the disturbance caused by ice scour than would a less-diverse community. With the exception of Stephenson and Stephenson's (1954) and Ellis and Wilce's (1961) description of intertidal zonation patterns in northern Canada, little is known about the effects of scouring by ice on rocky shores. We focus on the Pribilof Islands because the intertidal community there appears to be potentially very diverse. Moreover, these islands are near the southern limit of sea ice, and therefore the community there can be compared with those at ice-free islands at about the same latitude.

IV. STUDY AREAS

This report considers marine intertidal communities primarily in three coastal regions of Alaska: (1) Bristol Bay, (2) the Pribilof Islands, and (3) Norton Sound. Two sites (Nukshak Island and Spectacle Island) in the western Gulf of Alaska which were not included in our previous reports are also included here to complete the record of our intertidal reconnaissance studies.

We established study sites on six types of beaches based on predominant substrate composition. Maps of the distribution of substrate types in the three coastal regions considered here are contained in Sears and Zimmerman (1977). The types of substrate and percentages of our study sites on beaches with each type of substrate are as follows: boulder, 27%; sand, 27%; bedrock, 22%; mud, 17%; gravel, 4%; peat, 2%. Zimmerman et al. (1977) show the percentage of major intertidal substrate types for the three coastal regions we consider here. Comparing the latter percentages with the distribution of substrate types among our sites showed that we sampled substrate types roughly in proportion to their occurrence, although we tended to oversample predominantly boulder beaches and undersample gravel beaches. The relative proportion of beach types and the distribution of substrate types among the sites in each coastal region and a more detailed physical description of each site are included in the introductions to each coastal region below.

V. SOURCES, METHODS, AND RATIONALE OF DATA COLLECTION

A. Field methods

We used four sampling methods in this study. The one used most extensively employed belt transects sampled systematically with 1/16 m^2 quadrats. Transects were laid roughly perpendicular to the shoreline, usually from the level of mean high water or above the water's edge at low tide. The number of transect lines at each site and the sampling interval on each line depended on the slope, width, and topography of the beach.

At two sites (Cape Lapin and Eider Point) we used randomly sampled belt transects. These transects were laid parallel to the shoreline at arbitrarily selected tidal levels within major intertidal zones termed strata. We sampled them with a randomized nested design. They were subdivided into a set of contiguous 1-m^2 plots of which a subset was randomly selected for subsampling; each of these plots was subdivided into 1/16 m^2 quadrats two of which were sampled randomly. Three transects were laid at each site. These transects will hereafter be referred to as random transects to distinguish them from those described above which were sampled systematically.

The "arrow" sampling method (developed by R. Myren) was used primarily on vertical or near-vertical surfaces such as the sides of large boulders and rock outcrops. A facsimile of the area to be sampled and the general pattern of distribution of dominant organisms was sketched on a sheet of Mylar plastic. Numbered, uniformly-distributed dots were then placed on the sketch. The positions of a fraction (usually about 25%) of the dots were selected from a random number table. The locations on the rock surface corresponding to the randomly-selected dots were marked with numbered arrows. A quadrat (1/16 m^2) was then placed at the tip of each arrow, photographed, and its elevation determined (see below). The arrow site was not disturbed; plots of the same size with similar biological cover in a nearby area were sampled as described below.

At a few sites, transect sampling was supplemented by a method whereby a quadrat or corer was either tossed or placed on the beach haphazardly and sampled in the same way as the transect quadrats.

The procedure for sample collection on rocky shores was the same using all of the above methods. The area within each frame was photographed to record the coverage of larger organisms (visual estimates of this coverage were often made as well), and then the plot was scraped to bare rock and the organisms bagged and fixed in 10% formalin.

We used only transects and the haphazard sampling method at sites with unconsolidated sediments (mud, sand, gravel, peat). One-liter ($[10\text{ cm}]^3$) cores were taken. Most cores were collected from the surface of the substrate to 10 cm deep, but occasionally deeper (depth-range 10 to 20 cm) cores were also collected. Hereafter these depth ranges will be referred to as surface and deep, respectively. Exceptions to the above coring methods are as follows: (1) At our sites in Norton Sound 7 or 8 1-liter cores were frequently lumped into one sample or the sediment from one spot on the beach was shoveled into 7- or 8-liter buckets. In either case the samples were sieved on site through one or more screens, the finest having 2 mm openings, and then fixed for subsequent sorting. (2) At Cape Glazenap and Blaine Point in Izembek Lagoon, 1/16 m^2 quadrats were excavated to a depth of 5 and 7 cm respectively.

The elevations of samples taken by all of the above methods were determined with a transit and level rod using standard surveying techniques.

The reference level was the level of low tide predicted in the Tide Tables (Anonymous 1976).

In addition to taking quantitative samples, we made qualitative collections, and general observations regarding biological interactions and the distribution, relative abundance, and natural history of obvious organisms. Minor deviations from the procedures reviewed above are included in the descriptions of each site when appropriate.

B. Laboratory procedures

All samples were sorted by the Marine Sorting Center of the University of Alaska. All organisms were identified, counted (except organisms for which individuals could not be readily distinguished, e.g., many species of algae, sponges, bryozoans, etc.), and weighed (wet weight and dry weight). Organisms from most major phyla were identified to species. Invertebrates from the following taxa were not usually identified below the level of order by the Sorting Center: Porifera, Cnidaria, Platyhelminthes, Nemertea, Nematoda, Oligochaeta, Copepoda, Tanaidacea, Insecta, Arachnida, Acarina, Sipuncula, Bryozoa, and Ascidiacea. Counts and weights of mussels and limpets were recorded separately for two or three size categories. When most organisms had been removed and all that remained was a diverse mass of small fragments, estimates of the remaining individuals were determined by subsampling the residue.

C. Sources of bias and error

Only those samples collected on the random transects at Cape Lapin and Eider Point were strictly random; consequently, at most sites, we did not make a truly unbiased estimate of the variability in the abundance, biomass, or coverage of the organisms found there. When it was desirable to test specific hypotheses statistically and sample sizes were large enough, we chose at random a subset of the set of samples taken at each site.

The sample surface (i.e. the area of substrate enclosed by each quadrat or core) was small (1/16 m^2 or 100 cm^2) but probably adequate for sampling most species including the dominant competitors for space in the intertidal region. However, the sample size was clearly too small to adequately sample large predators such as starfish. Therefore, our data cannot be used to estimate the abundance of these species.

Other sources of bias and error that affect the usefulness of the data from particular regions or sites are included in the Results section.

VI. RESULTS

A. Distributions and Relative Abundances of Intertidal Biota

1. Bristol Bay and Eastern Aleutian Islands

We sampled 16 sites in Bristol Bay and the eastern Aleutian Islands (Fig. 1). About half of the beaches sampled were sandy or muddy, and half were rocky; pertinent sampling information for these sites is contained in Table 1.

In addition to the above sampling activity, S. Zimmerman and J. MacKinnon conducted a reconnaissance of the littoral region of 14 sites in eastern Bristol Bay from the Nushagak Peninsula to Port Heiden from 29 August to 5 September 1976. Zimmerman and Merrell (1976) list their observations.

From 6 to 12 September 1976, MacKinnon accompanied Drs. A. Salenger, B. Hunter, and J. Dingler of the U. S. Geological Survey on a reconnaissance of 39 sites on the north coast of the Alaska Peninsula from Kvichak Bay to Unimak Island. His observations are summarized in Zimmerman and Merrell (1976).

Also included here are descriptions of intertidal communities at two sites, Nukshak and Spectacle Islands, in the western Gulf of Alaska (Fig. 1) which were not included in our previous reports. Lists of species of intertidal plants and invertebrates identified in samples from all sites in this section are contained in Appendices IIA and IIB.

1.1 Nukshak Island (Cape Nukshak)

Nukshak Island is in Shelikof Strait just off Cape Nukshak on the Alaska Peninsula (Figs. 1 and 2*; Table 1). The island is small (0.9 km long, 40.5 m high [U.S. Coast Pilot 1964]) with a bedrock intertidal area rising to two

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* All maps of specific sites are modified from Sears and Zimmerman (1977; see List of Figures).

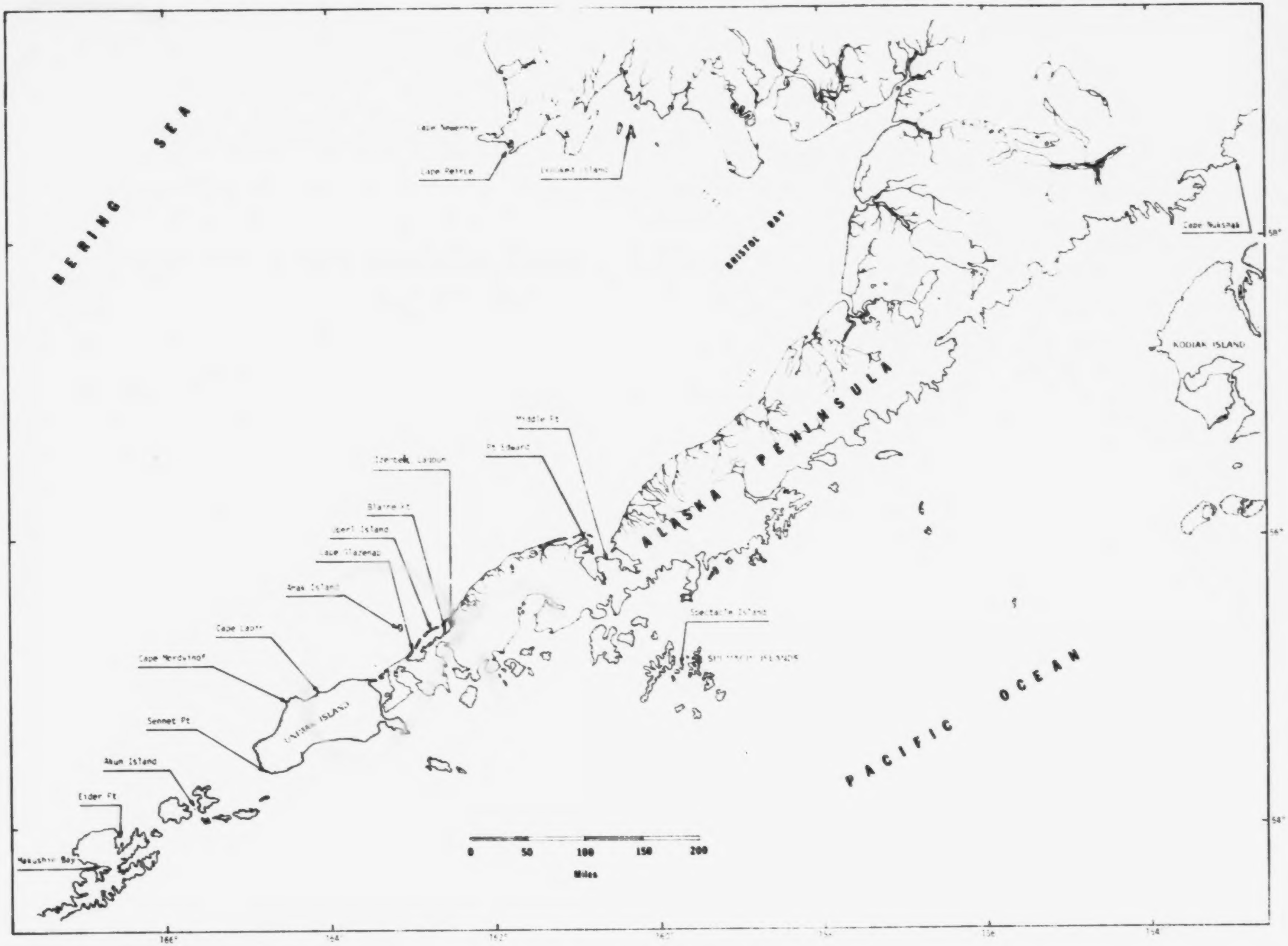
BERING SEA
Cape Newenham
Cape Peirce
Crooked Island
BRISTOL BAY
Cape Nukshak
58°
KODIAK ISLAND
ALASKA PENINSULA
Middle Pt
Pt Edward
Izembek Lagoon
Blaine Pt
Operl Island
Cape Glazenap
Amak Island
Cape Lapin
Cape Mordvinof
UNIMAK ISLAND
Sennet Pt
Akun Island
Eider Pt
Makushin Bay
Spectacle Island
SHUMAGIN ISLANDS
56°
PACIFIC OCEAN
54°
0
50
100
150
200
Miles
166°
164°
162°
160°
158°
156°
154°

Fig. 1. Location of sites in the western Gulf of Alaska, Bristol Bay and on the Aleutian Islands.

Table 1. Pertinent sampling information for 18 sites in the western Gulf of Alaska, Bristol Bay, and the Aleutian Islands.

Site Name	Coordinates Lat.(N)	 Long.(W)	Substrate	Dates sampled	Tidal Range sampled (cm)	Sampling method(s)[a]	Sample Surface area or volume(ℓ.)[b]	Sample Size[c]
Western Gulf of Alaska								
Nukshak Island	58°23'	153°59'	bedrock	23,24 May 1975	-20 to 250	Transect(2),arrow	1/16 m^2	18(T), 8(A)
				8 August 1975	70 to 310	Transect(2),arrow	1/16 m^2	22(T), 8(A)
Spectacle Island	55°07.2'	159°44.6'	boulder/bedrock	25,26 May 1975	-18.3 to 195.1	Transect(2),arrow	1/16 m^2	21(T), 12(A)
				11 August 1975	24.4 to 140.2	Transect(2),arrow	1/16 m^2	22(T), 4(A)
Southern Bristol Bay								
Middle Point	55°51.7'	160°40.0'	mud/sand	23 July 1975	not recorded	Haphazard	1	6
Point Edward	55°59.5'	160°51.6'	mud/sand	22 July 1975	-42 to 104	Transect	1	38
Moffet Lagoon	55°26.0'	162°31.0'	mud	15 June 1976	-12.2 to 57.9	Transect	1	15
Blaine Point	55°22.3'	162°39.7'	mud	15 June 1976	-9.1 to 82.3	Transect	4.4	11
Izembek Lagoon	55°18.9'	162°45.5'	mud	16 June 1976	6.1 to 76.2	Transect,selected	6.25, 1	11(T), 4(S)
Operl Island	55°23.5'	162°47.0'	sand	16 June 1976	not recorded	Haphazard	1	2
Cape Glazenap A	55°13.2'	163°02.0'	mud/sand	16 June 1976	not recorded	Haphazard	1	2
Cape Glazenap B	55°12.8'	163°01.2'	mud/sand	16 June 1976	-9.1 to 30.5	Transect,haphazard	3.1, 1	10(T), 3(H)
Amak Island	55°24.1'	163°09.3'	boulder/bedrock	19 July 1975	-27 to 223	Transect	1/16 m^2	20
Cape Lapin	54°56.7'	164°08.0'	rubble to boulder	17 June 1976	70.1 to 317.0	Transect(4), nested quadrat	1/16 m^2	24
Cape Mordvinof	54°55.8'	164°26.8'	bedrock/boulder/ rubble	24 July 1975	27.4 to 283.5	Transect,arrow	1/16 m^2	7(T), 4(A)
Sennet Point	54°29.1'	164°54.4'	bedrock/boulder/ rubble	8 June 1976	6.1 to 368.8	Transect	1/16 m^2	7
Aleutian Islands								
Akun Island	54°08.5'	165°38.7'	bedrock	18 July 1975	-21 to 140	Transect,arrow, nested quadrat	1/16 m^2	20(T), 4(A)
Eider Point	53°57.5'	166°35.1'	rubble	14 June 1976	-15.2 to 216.4	Transect(4), nested quadrat	1/16 m^2	30
Portage Bay	53°44.0'	166°45.8'	bedrock	25 July 1975	80 to 161	Transect,arrow	1/16 m^2	14(T), 7(A)
				13 August 1975	67 to 158	Transect	1/16 m^2	22
Northern Bristol Bay								
Crooked Island	58°39.5'	160°16.5'	bedrock	20 July 1975	1 to 226	Transect	1/16 m^2	18
Cape Peirce	58°34.4'	161°45.5'	bedrock	21 July 1975	28 to 216	Haphazard,arrow	1/16 m^2	13(H), 4(A)

a. Numbers in parentheses indicate number of transects sampled per site, if more than one; b. Numbers not followed by m^2 designations are liter measurements; c. Letters in parentheses refer to sampling method: T = Transect; A = Arrow; S = Selected; H = Haphazard.

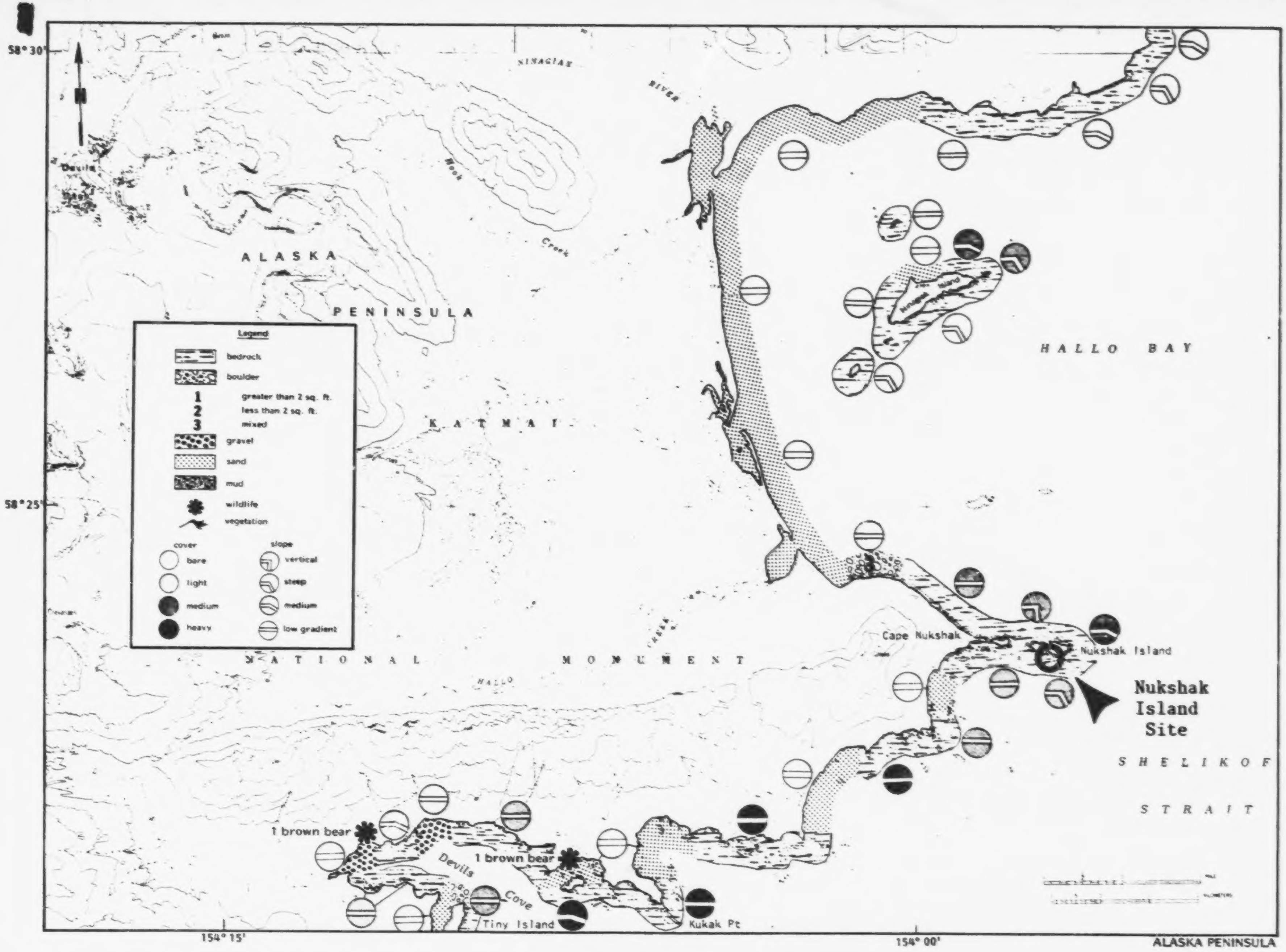
58° 30'
58° 25'
154° 15'
154° 00'
ALASKA PENINSULA
NINAGIAK RIVER
Hook Creek
ALASKA PENINSULA
KATMAI NATIONAL MONUMENT
HALLO
HALLO BAY
Ninagiak Island
Cape Nukshak
Nukshak Island
Nukshak Island Site
SHELIKOF STRAIT
1 brown bear
1 brown bear
Devils Cove
Tiny Island
Kukak Pt
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 2. Location of site at Nukshak Island.

grass-covered knolls. Our survey site was on the south side of the island. The bedrock was hard, granite-like, and where we sampled, gently sloping (Fig. 3). Outside the area we sampled in May 1975, we observed low-gradient bedrock with fissures and depressed areas filled with water. In August 1975, we laid Transect 2 parallel to the water's edge and perpendicular to Transect 1 at the level of meter 12 in order to sample the low-gradient area.

Barnacles were the dominant organism at Nukshak Island. Four species, *Balanus glandula*, *B. balanoides*, *B. cariosus*, and *Chthamalus dalli* were found although *B. balanoides* was not identified from our May 1975 samples. Table 2 shows the frequency of occurrence of selected species along transects in May and August 1975. The predaceous snails *Nucella lima*, *N. lamellosa*, and *N. canaliculata* were common, but they exhibited gregarious behavior and were inadequately sampled by the transects. *Nucella lima* was observed feeding on *Balanus* sp. and many empty tests were present, apparently from predation by *Nucella*. In August 1975, Transect 2 intersected a low-gradient area containing some standing water and many crevices; *Leptasterias hexactis* was collected in every quadrat along the transect. The large chiton *Katharina tunicata* was abundant throughout the low intertidal zone at Nukshak Island; however, it was under-represented in our quadrat collections. *Mytilus edulis* occurred in many of our quadrats, but its distribution was patchy and it was not abundant. *Littorina sitkana* was very abundant in the high intertidal zone where it was often observed in clusters, grazing on a filamentous green alga.

Macrophyte cover was light, except in the low intertidal zone where *Alaria* spp. were abundant. *Fucus distichus* was collected in most higher-elevation quadrats but most individuals were small. *Porphyra* spp. were

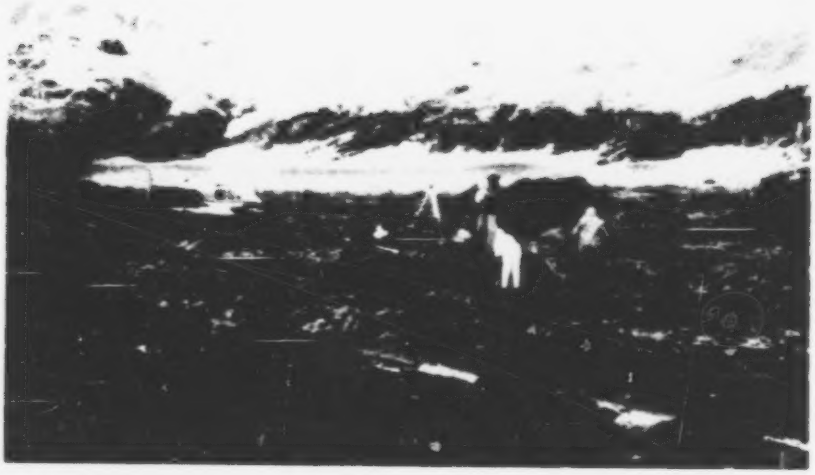

Fig. 3. Nukshak Island sampling area. Site of Transect 1; Transect 2 is off photo to right.

Table 2. Frequency of occurrence of selected species along four transect lines on Nukshak Island, May and September 1975.

	May 1975		August 1975	
Species	Transect 1 (2.2 to 1.1m) (n = 7)	Transect 2 (2.5 to -0.2m) (n = 11)	Transect 1 (3.1 to 0.8m) (n = 17)	Transect 2 (1.4 to 1.0m) (n = 5)
Fucus distichus	.86	.45	.82	.60
Littorina sitkana	1.00	.55	1.00	.80
Balanus glandula	1.00	.55	.65	.40
Balanus balanoides	0	0	.47	.20
Balanus cariosus	.29	.64	.53	1.00
Chthamalus dalli	.43	.64	.35	.60
Mytilus edulis <1.5 cm	.71	.55	.76	1.00
Mytilus edulis 1.5-2.0 cm	.71	.18	.24	.60
Mytilus edulis >2.0 cm	.71	.09	.18	.40
Collisella pelta	.86	.55	.71	1.00
Nucella spp.	.29	.09	.35	.80
Leptasterias hexactis	0	.45	.24	1.00
Turtonia occidentalis	.71	.36	.47	.40
Porphyra sp.	.67	.45	.35	.20
Alaria spp.	.14	.45	.24	.20
Polychaetes	1.00	1.00	1.00	1.00
Oligochaetes	1.00	.64	.88	1.00
Flatworms	.71	.45	.59	.40
Pagurus hirsutiusculus	0	0	.18	1.00
Katharina tunicata	0	.36	0	.40

collected in both May and August (many species of *Porphyra* occur only in the spring) at the high and mid-intertidal levels. Encrusting and foliose coralline algae were very abundant in tide pools. Clumps of *Mytilus edulis* and *Modiolus modiolus* growing in the tide pools were almost completely covered with encrusting coralline algae. *Laminaria* sp. was also common in tide pools and on a steep rock face seaward from the end of the transects; conditions were too hazardous for collections to be made here.

1.2 Spectacle Island, Shumagin Islands

Spectacle Island is a small rocky island with grass-covered slopes which lies between Nagai and Big Konuiju Islands (Figs. 1 and 4; Table 1). Our sampling site was located on a small islet off the southwest shore of Spectacle Island; a gravel spit connects these two bodies. The substratum was bedrock overlain by boulders (Figs. 5 and 6).

The Spectacle Island site has a fairly heavy cover of macrophytes, with a predominance of grazers and filter feeders in its invertebrate population. We found 29 species of algae along the transects in May and August. Although the greatest number of algal species present were rhodophytes, *Fucus distichus* (a phaeophyte) had the greatest biomass. Polychaetes contributed the greatest number of invertebrate species, with 23 species found in May and 18 species present in August. Grazers, including pulmonates, limpets, other gastropods, and chitons, were abundant. We found 18 species of grazers in May and 13 species of grazers in August. Several barnacle and mussel species were found during both sampling periods, but in small numbers. We found few predators. A single individual of the carnivorous gastropod species *Nucella lamellosa* was collected in May; none were found in August. A few small sea stars, *Leptasterias hexactis*, were collected in both May and August. *Nereocystis luetkeana*, a floating kelp,

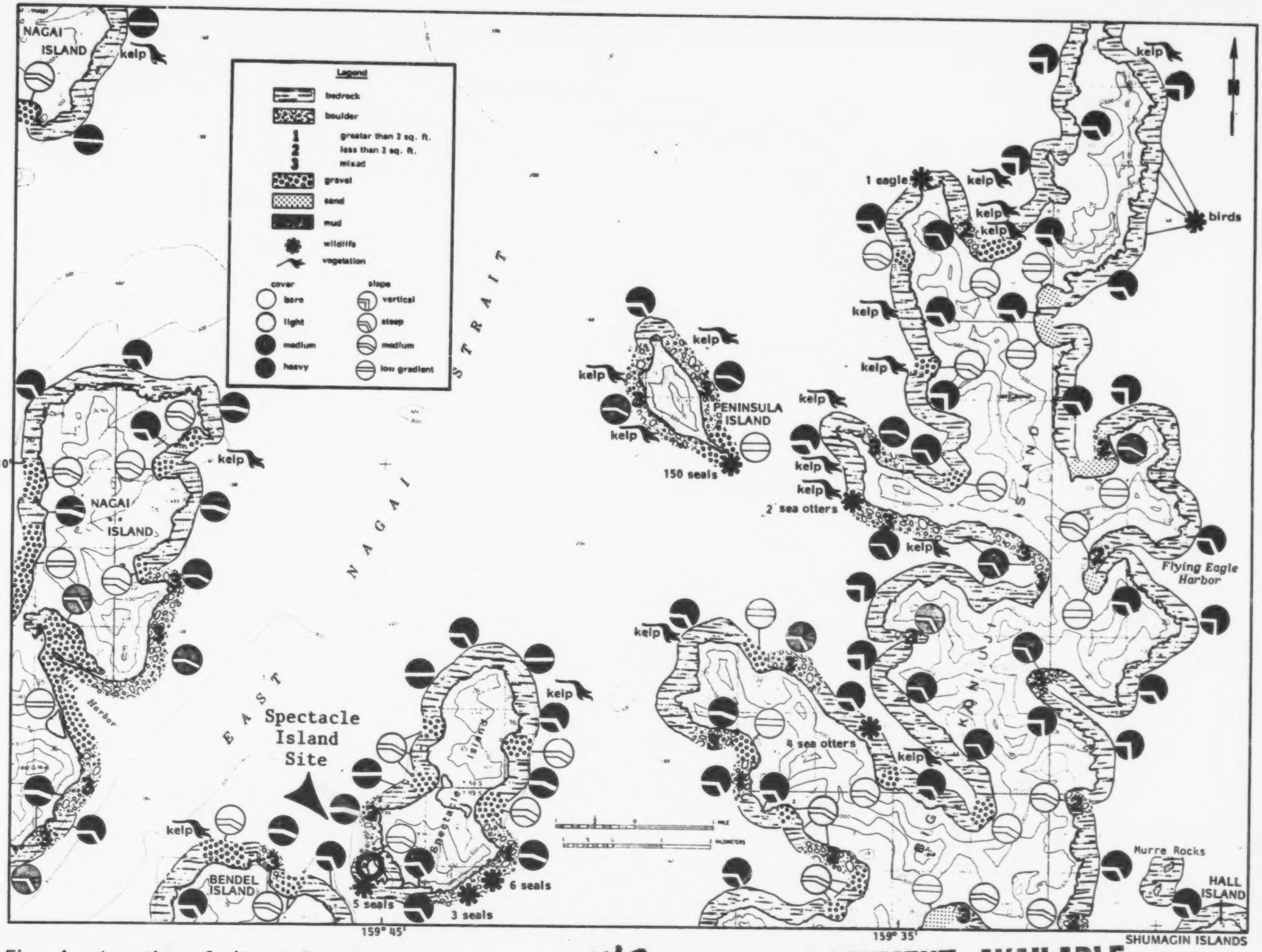
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
NAGAI ISLAND
kelp
55° 10'
NAGAI ISLAND
EAST NAGAI STRAIT
Spectacle Island Site
BENDEL ISLAND
5 seals
3 seals
6 seals
PENINSULA ISLAND
150 seals
1 eagle
birds
2 sea otters
4 sea otters
Flying Eagle Harbor
BIG KONIUJI ISLAND
Murre Rocks
HALL ISLAND
159° 45'
159° 35'
SHUMAGIN ISLANDS

Fig. 4. Location of site at Spectacle Island.

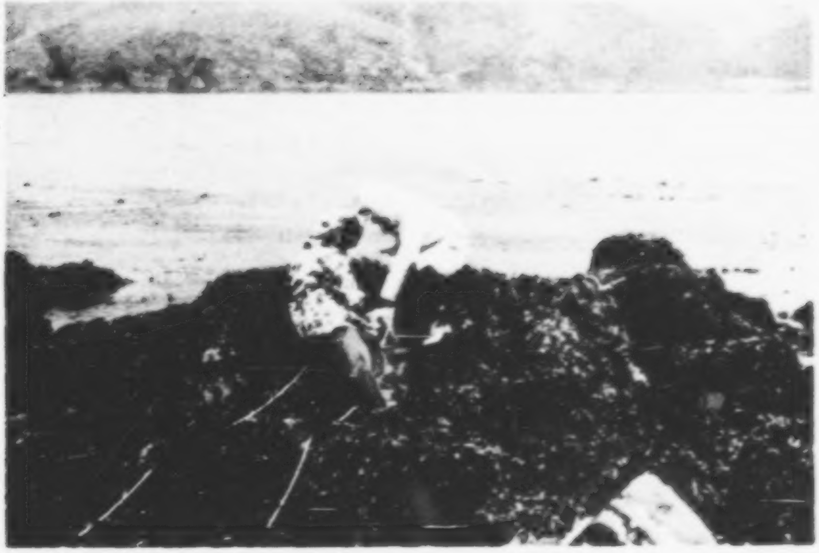

Fig. 5. Replicate transect lines at Spectacle Island site, August 1975. Note floating kelp offshore.

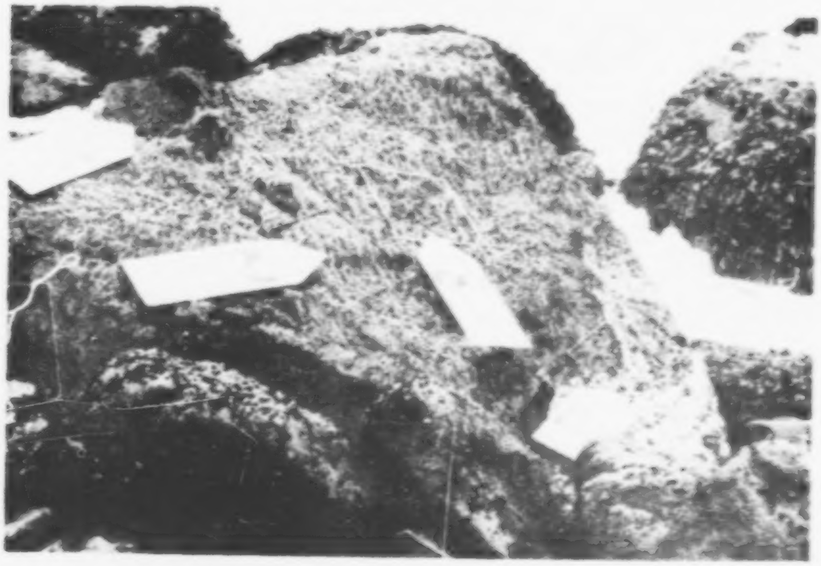

Fig. 6. Arrows in place to study abundance and distribution of organisms within zones at Spectacle Island, Alaska.

was abundant offshore (Fig. 5). During both sampling periods we saw sea otters swimming among the kelp. Although we saw only a few sea otters when we worked at the site, they are present throughout the area (Sears and Zimmerman 1977). Sea otters may help to account for some of the conditions we observed such as ungrazed algal beds, increased algal competition for space, and reduced populations of sessile invertebrates (Palmisano and Estes 1977).

1.3 Middle Point, Port Moller

Middle Point is located along the western shore of Port Moller (Figs. 1 and 7; Table 1). Our site consisted of an extensive tidal flat covered with the eelgrass Zostera marina. The substrate was mud overlying sand and the declination was so slight that one could walk seaward for hundreds of meters without encountering any change in water depth; even at low tide water remained on the flat to a depth of about 3 to 5 cm.

One of the most numerous groups or organisms at the site were caprellid amphipods which clung to the eelgrass. We estimated that there were an average of five caprellids per square centimeter throughout the flat; one sample with particularly heavy eelgrass cover yielded an actual count of 1234 caprellids per 100 cm^2. Polychaetes were another important group of invertebrates in our samples with 32 species in 18 families represented, although no single species was especially numerous. We collected five species of bivalves at this site; none was particularly numerous except Macoma balthica which was abundant in one sample collected on the highest part of the beach, above the eelgrass.

Middle Point is probably a very productive site. Eelgrass beds are generally highly productive marine systems (for recent reviews see Phillips 1974 and McRoy and McMillan 1977). The rhizomes of eelgrass stabilize the

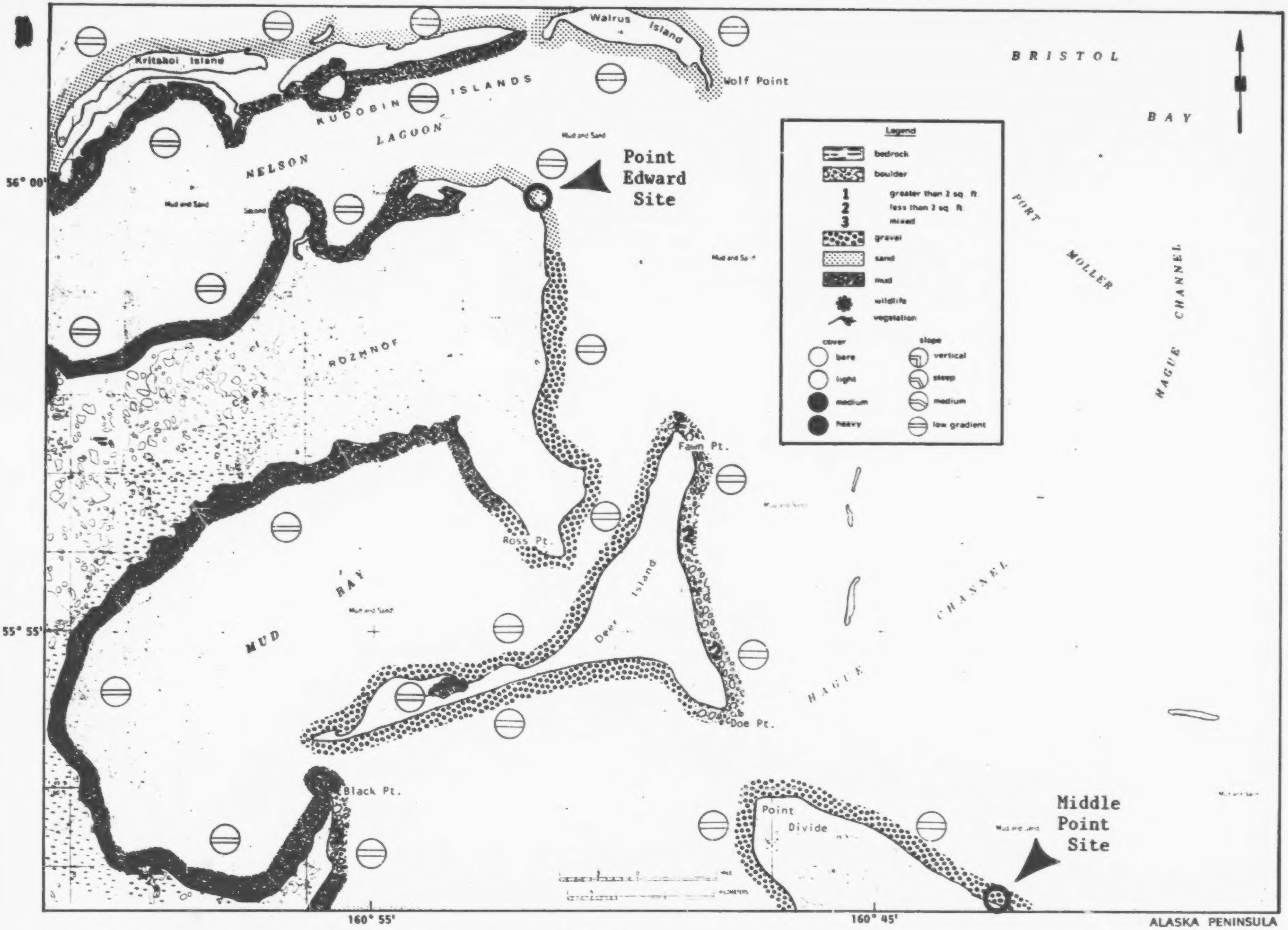
Kritskoi Island
Walrus Island
Wolf Point
KUDOBIN ISLANDS
NELSON LAGOON
BRISTOL BAY
Point Edward Site
PORT MOLLER
HAGUE CHANNEL
ROZHNOF
Fawn Pt.
Ross Pt.
Deer Island
Doe Pt.
MUD BAY
Black Pt.
Point Divide
Middle Point Site
56° 00'
55° 55'
160° 55'
160° 45'
Mud and Sand
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
ALASKA PENINSULA

Fig. 7. Location of sites at Point Edward and Middle Point.

146

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substrate and, upon decay, the eelgrass yields detritus which nourishes the benthic community; it also provides physical support and protection for many small organisms such as caprellids. Although this area was not observed during high tide, the abundant food supply and protective cover afforded by the eelgrass probably provide a valuable habitat for juvenile fish.

1.4 Point Edward, Port Moller

Point Edward is at the northeastern end of Cape Rozhof near the entrance of Port Moller (Figs. 1 and 7). We collected 1-liter samples on a broad, gently-sloping, mud/sand beach (Table 1, Fig. 8); all cores taken along our transect were below Mean Tide Level (MTL) and over half (60%) were below Mean Lower Low Water (MLLW).

Most of the species in our samples were polychaetes with at least 29 species present. The Class Bivalvia was represented by six species, other major classes of invertebrates by no more than three or four species. Diatoms, a green alga (Enteromorpha intestinalis), and two red algae (Porphyra sp. and Cryptosiphonia woodii) were the only algae in our samples.

Eight of the ten species of invertebrates having the greatest average wet weight in our cores were polychaetes (Table 3), but bivalves, especially Macoma balthica, made up most of the biomass. Although Macoma was outweighed by Mya arenaria in three cores and by Clinocardium nuttallii in one core, the latter two species were rare in our samples and Macoma was clearly the preponderant species (79%) in most cores.

Among the Polychaeta, sedentary forms constituted about 75% of those species showing the greatest average wet weight in our samples and 70% of all polychaetes present. Nephtys caeca occurred more frequently than any other species, but Arenicola gracilis had a substantially higher median abundance and ranked just below Macoma in average biomass.

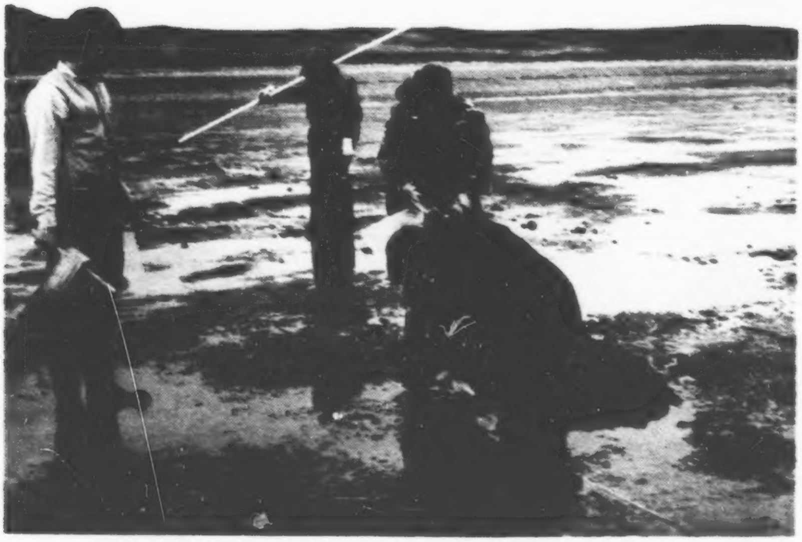


Fig. 8. Core sampling at Pt. Edward, Port Moller, Alaska Peninsula. 22 July 1975.

Table 3. Statistics for the ten species of invertebrates showing the greatest average rank on the transect at Point Edward, Port Moller.

Species	Average rank[a]	Wet weight (g)[b]	Frequency of occurrence[c]	Abundance[d]	Dominance[e]
Macoma balthica	11.5	5.1, 0.2-18.8	0.92	17, 3-48	30/38
Arenicola gracilis	7.5	0.2, 0.001-1.6	0.71	19, 1-44	1/38
Axinopsida serricata	7.2	0.03, 0.002-0.1	0.87	6, 1-36	0
Nephtys caeca	6.6	0.02, 0.001-1.8	0.95	4, 1-11	0
Spio filicornis	6.3	0.02, 0.001-0.2	0.87	4, 1-15	0
Capitella capitata	4.7	0.05, 0.001-0.7	0.55	2, 1-17	1/38
Owenia fusiformis	3.6	0.03, 0.003-0.1	0.40	2, 1-4	0
Pholoe minuta	3.4	0.005, 0.001-0.02	0.82	3, 1-12	0
Ampharete arctica	3.4	0.004, 0.001-0.05	0.68	1, 1-7	0
Polycirrus medusa	3.2	0.07, 0.001-0.6	0.29	2, 1-6	0

a. Rank of wet weight of each species averaged over 38 cores (1, lowest weight). Species listed in order of decreasing rank.
b. Median and range of wet weights per liter in all cores where the species was found.
c. Proportion of quadrats that contained the species.
d. Median and range of number of individuals per liter in all cores where the species was found.
e. Proportion of cores in which the species was among those making up 50% of the wet weight. Summation in each core began with the heaviest species.

1.5 Moffett Lagoon

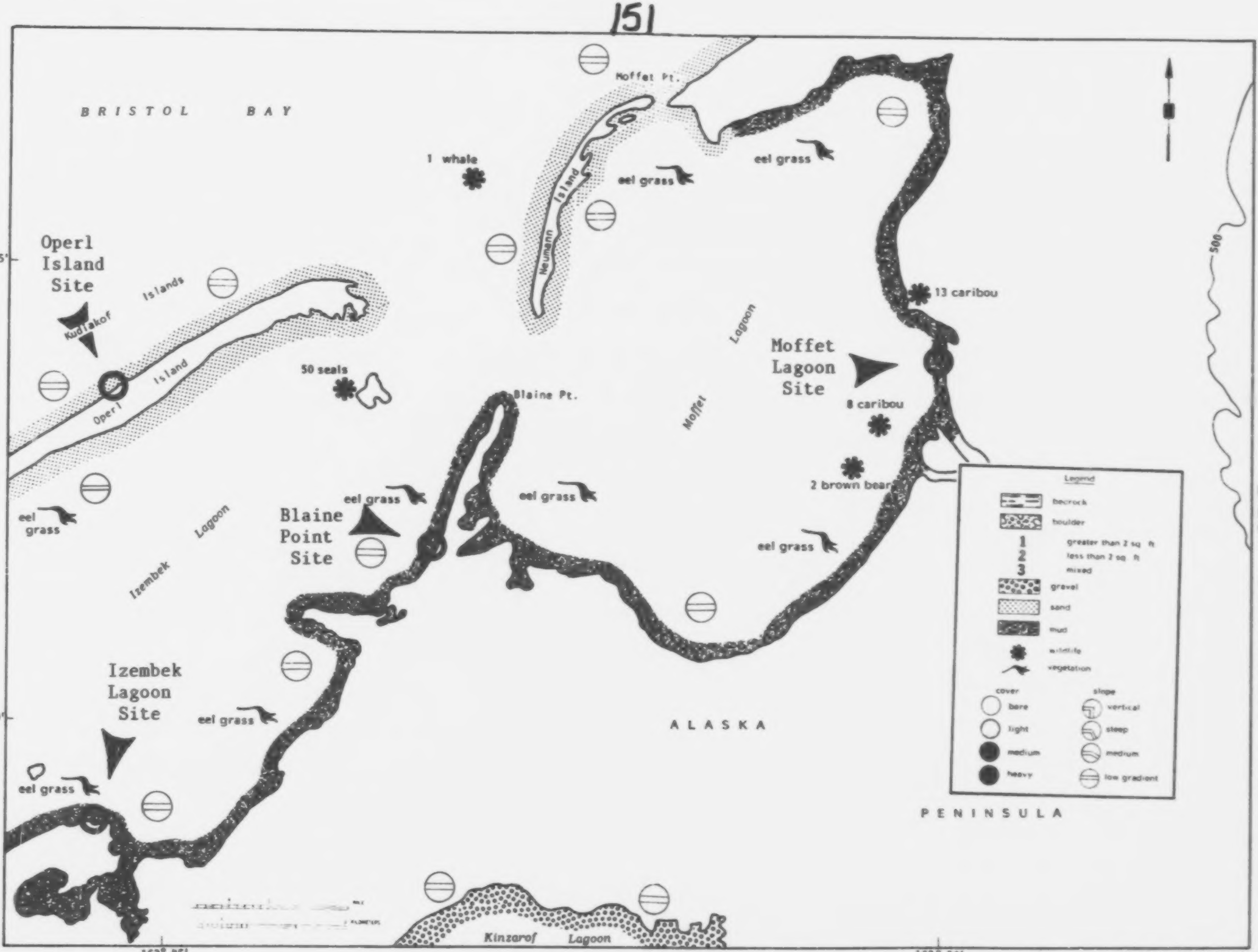
The Moffet Lagoon site located north of Izembek Lagoon was a low-gradient mudflat facing west (Figs. 1 and 9; Table 1). The substrate was composed of silty sediment with little eelgrass or other vegetation present. The entire area for a considerable distance around the site was similar with respect to an absence of eelgrass. Elevations ranged from 0.58 meters at the shoreline station to -0.12 meters at the most seaward sampling station.

The scarcity of biota expressed as biomass wet weight per 100 cm^2 sample area is summarized in Fig. 10. The most numerous species was *Rhynchospio* sp. of which a total of 51 individuals was found in all the samples. The numbers of individuals of other species included 7 *Edotea* sp., 5 *Pontoporeia affinis*, 1 *Laonice cirrata*, 11 nematodes, 4 oligochaetes, 1 Lysianassid, 1 nemertean, 1 Maldanid fragment, and one unidentified amphipod.

Most of the biota was found in the 0 to 3 cm stratum of the core sample, although *Rhynchospio* sp. was nearly equally divided between the 0 to 3 cm and the 3 to 6 cm deep strata in the samples. Some contamination of the sample may have occurred because two individuals of the genus *Pontoporeia*, normally shallow burrowers, were found in the deepest (6 to 9 cm) stratum.

1.6 Blaine Point

Blaine Point lies at the tip of the peninsula separating Izembek and Moffet Lagoons (Figs. 1 and 9). We collected eleven 4.4-liter cores on a 152 m long transect in an eelgrass bed (Fig. 11, Table 1). All but one core were taken below MTL; 55% were taken below MLLW. Two of the 11 cores collected were poorly fixed and were not used in the analysis.


BRISTOL BAY
Moffet Pt.
Neumann Island
1 whale
eel grass
eel grass
eel grass
Operl Island Site
Kudiakof Islands
Operl Island
50 seals
Blaine Pt.
Moffet Lagoon
Moffet Lagoon Site
13 caribou
8 caribou
2 brown bear
eel grass
eel grass
eel grass
Blaine Point Site
Izembek Lagoon
eel grass
Izembek Lagoon Site
eel grass
eel grass
ALASKA
PENINSULA
Kinzarof Lagoon
55° 25'
55° 20'
162° 45'
162° 30'
500
Legend
bedrock
boulder
1 greater than 2 sq. ft
2 less than 2 sq. ft
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
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steep
medium
low gradient
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Fig. 9. Location of sites at Operl Island, Izembek Lagoon, Blaine Point, and Moffet Lagoon.

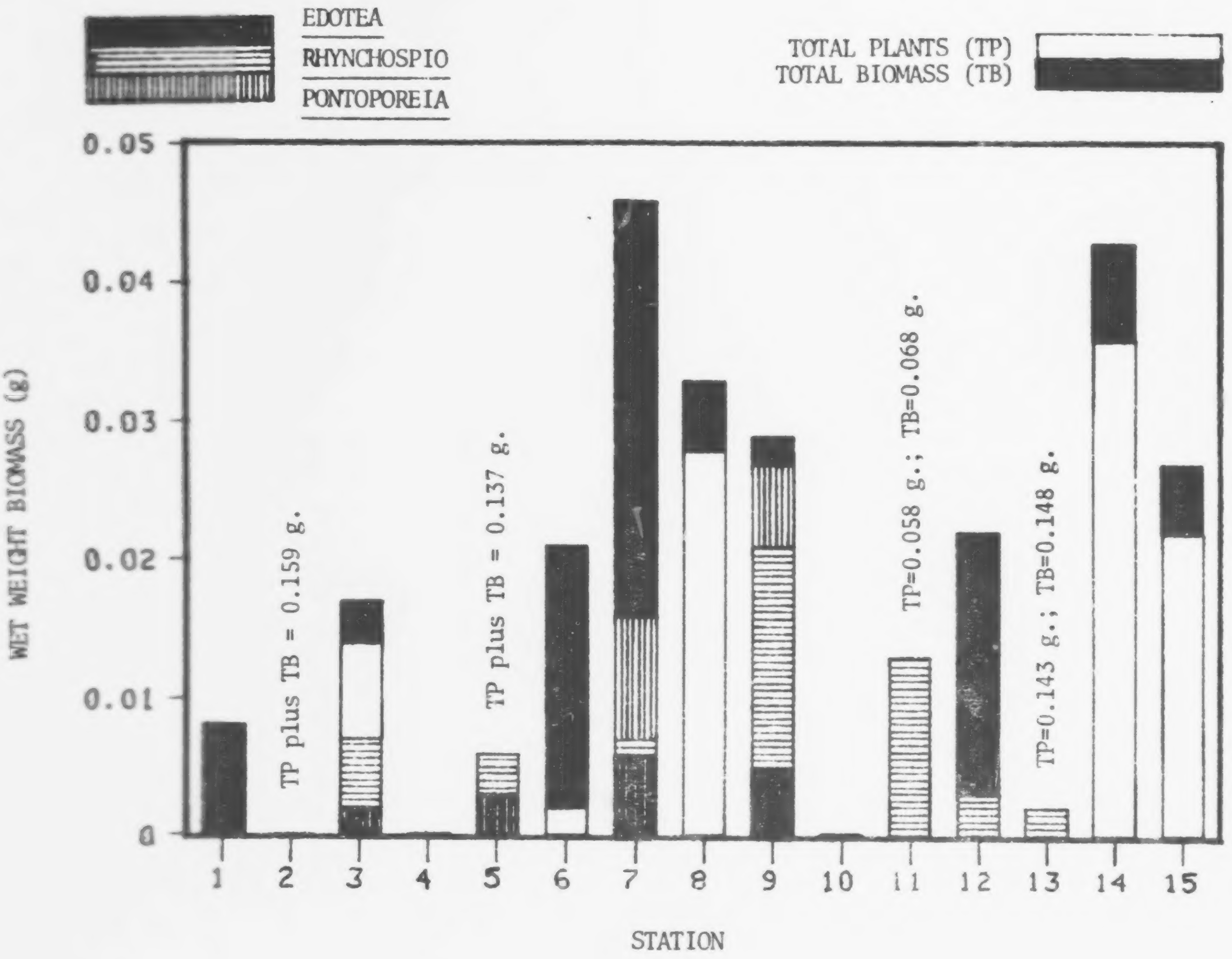

Fig. 10. Biomass in core samples at Moffet Lagoon. 15 June 1976.

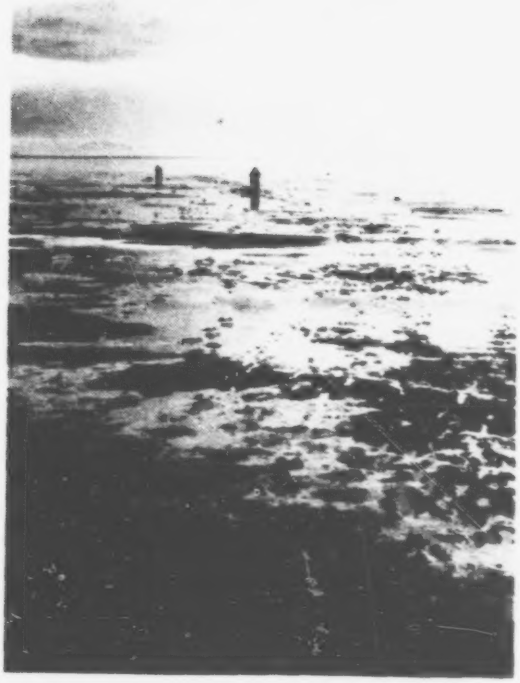

Fig. 11. Blaine Point site. 15 June 1976.

Potamogetonaceae (presumably Zostera marina) occurred in all samples and showed the greatest wet weight (Table 4) in all but one sample (at the lowest tidal level) in which Macoma brota was preponderant.

The number of species in our samples averaged 31. Polychaetes constituted an average of 60% of the species present in each plot; at least 43 species of polychaetes were represented in our samples (Appendix II-B). Nine of the ten plant or animal species with the greatest average wet weight (as indicated by average rank) in Blaine Point samples were polychaetes; five of these species were present in all samples (Table 4).

Next to Polychaeta, Bivalvia and Gastropoda were the most diverse groups with five and four species, respectively, found in our samples (Table 4). When they occurred, bivalves and gastropods usually ranked high in wet weight, but no species belonging to these groups occurred in more than three samples. Unidentified bivalves and gastropods without shells were present in all samples.

Other major phyla of invertebrates were never represented by more than two species. Among the algae only Ahnfeltia sp., A. plicata, and unidentified red and brown algal fragments were present.

1.7 Izembek Lagoon

The Izembek Lagoon site was on a low-gradient mud flat with a northern exposure (Figs. 1 and 9; Table 1). The substrate was composed of silt and mud covered with eelgrass (approximately 80%).

One transect running approximately north was established normal to the shoreline. Nine 10 cm^3 (1 ℓ) cores were taken, one every 15 meters seaward to 135 meters; one additional sample was taken near the water's edge, 205 meters horizontal distance along the transect. Four selective core samples were removed in areas with little or no eelgrass cover at elevations of 0.77 m,

Table 4. Statistics for the ten species of plants and invertebrates showing the greatest average rank at the transect at Blaine Point, Izembek Lagoon.

Species	Average rank[a]	Wet Weight(g)[b]	Frequency of occurrence[c]	Abundance[d]	Dominance[e]
Potamogetonaceae	33.2	318, 3.6-539.8	1	-	8/9
Haploscoloplos panamensis	28.6	0.3, 0.02-1.3	1	37, 3-94	0
Praxillella affinis	27.0	0.6, 0.04-2.5	0.89	46, 17-117	0
Anaitides maculata	22.2	0.05, 0.03-0.4	1	5, 1-17	0
Owenia fusiformis	20.9	0.07, 0.002-0.4	1	5, 1-26	0
Harmothoe imbricata	20.4	0.3, 0.06-0.5	0.78	9, 1-12	0
Nephtys caeca	19.6	0.5, 0.02-1.3	0.67	4, 2-6	0
Capitella capitata	19.1	0.05, 0.01-0.1	0.89	54, 9-155	0
Eteone longa	16.2	0.03, 0.003-0.07	1	7, 1-16	0
Pholoe minuta	14.8	0.02, 0.002-0.06	1	6, 1-35	0

a. Rank of wet weight of each species averaged over nine cores (1, lowest weight). Species listed in order of decreasing rank.
b. Median and range of wet weights per liter in all cores where the species was found.
c. Proportion of quadrats that contained the species.
d. Animals only; median and range of number of individuals per liter in all cores where the species was found.
e. Proportion of cores in which the species was among those making up 50% of the wet weight. Summation in each core began with the heaviest species.

0.68 m, 0.55 m, and 0.06 m. In the analyses, mean number of species (density/100 cm^2) or wet weight biomass per station and the standard deviation of the mean were computed for selected species and expressed as number (density) or weight per 100 cm^2 area ± standard deviation of mean.

The elevations along the transect gradually decreased from the high to upper low intertidal zones, except for a slight elevation increase at station 6 (Fig. 12). The transect gradient was noticeably steeper for the two low intertidal stations, 10 and 11. Station 10 (elevation 0.37 m) was omitted from the analysis and figures because of poor preservation of the sample.

Eelgrass biomass varied in the higher elevation stations from a low value at station 7 (containing 1 g) to intermediate values at stations 1 and 3, and higher values at stations 2, 4, 5, 6 (Fig. 12). Eelgrass biomass was highest at the more seaward stations, station 8 (389 g) and station 11 (297 g; Fig. 12). At station 9, eelgrass biomass was similar to the moderate values of the higher elevation stations.

The community at the site was composed primarily of polychaetes, one or two species of pelecypods, and eelgrass (Fig. 13). A small unidentified pelecypod contributed significantly to the animal biomass at every station. Macoma balthica was found only at one station. The polychaetes dominating the animal biomass at most stations were Haploscoloplos panamensis, Harmothoe imbricata, Nephtys sp., and Praxillella affinis. A stalked sea anemone contributed to the biomass significantly and was present at six stations. Species not shown in Figure 13 which contributed large proportions of the biomass at single stations were Telmessus cheiragonus, an unidentified amphipod, and a stalked tunicate and anemone.

The most numerous species recorded at the site, present in every transect station but absent in two selected samples, was the unidentified

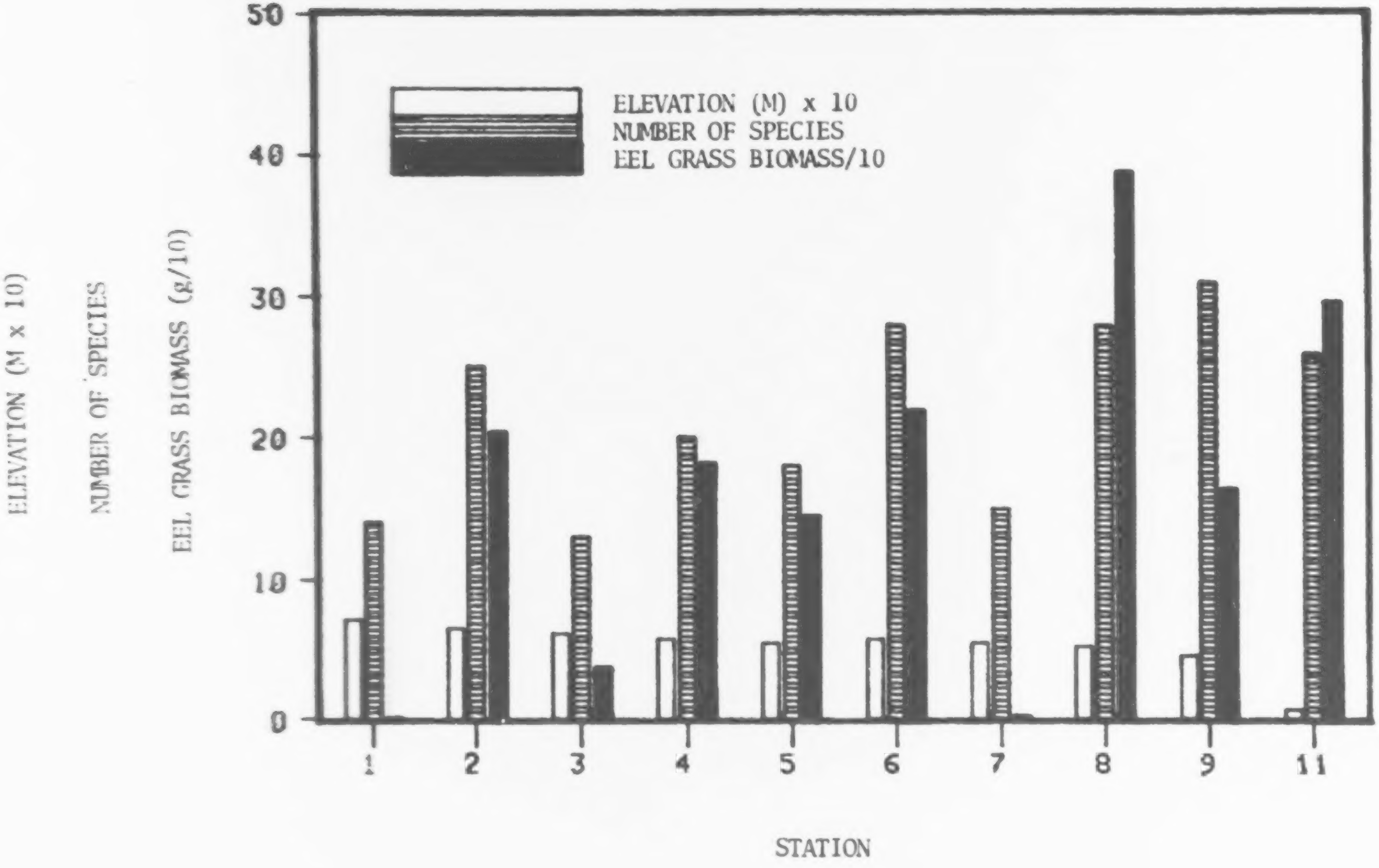

Fig. 12. Elevation, number of species, and eel grass biomass (wet weight) for the transect stations at Izembek Lagoon. 16 June 1976.

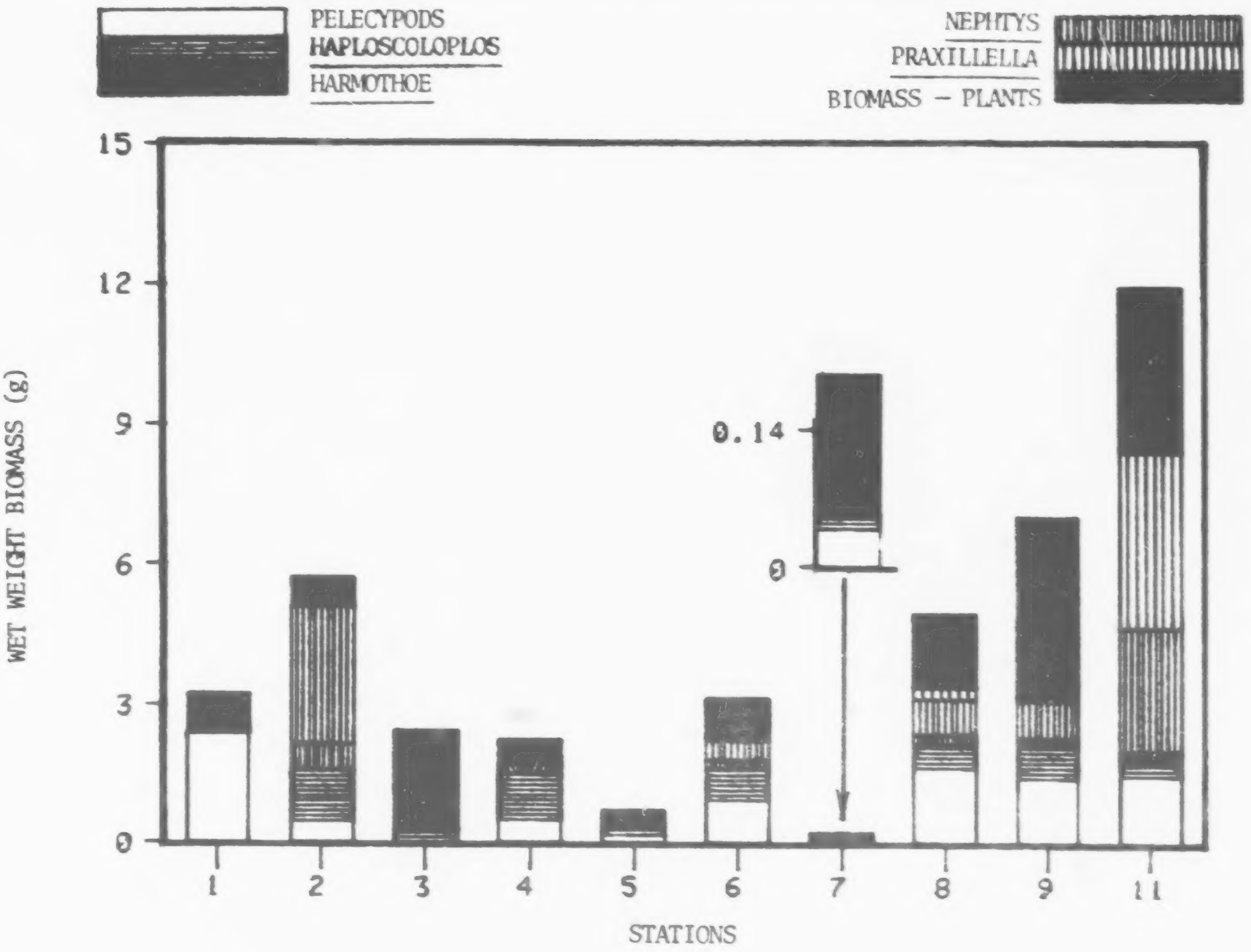

Fig. 13. Comparative biomass of dominant pelecypods and annelids found in transect stations at Izembek Lagoon. Total animal biomass is represented by biomass-plants (biomass minus plants).

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pelecypod with maximum count of 1,160 individuals at one station. An unidentified gastropod occurred in six stations along our transect and in two selected samples, with a peak count of 125 individuals. Animal biomass for the Izembek Lagoon site is shown in Figure 13 as biomass-plants (biomass minus plants) and ranged from 0.2 to 12 g per sample.

Polychaetes contributed the most species per taxonomic group at the site with approximately 36 species or groups represented. The most numerous single polychaete species at the site was Fabricia sabella with 219 individuals at an upper intertidal station (station 2); the same species was represented by only six individuals at one other station. Haploscoloplos panamensis was present in every coring sample except three (of 15) and the highest density of 51 individuals (average 21 ± 6) occurred at station 9, a low intertidal station. Capitella capitata was present in 10 samples with the highest density of 195 individuals (average 73 ± 25) also occurring at station 9. Other polychaetes with densities usually greater than 10 per station for at least one station included Praxillella affinis, Eulalia bilineata, Chaetozone setosa, Phyllodocids, Eteone longa, Sphaerosyllis pirifera, Nephtys sp., and Anaitides groenlandica.

Crustaceans were not abundant. Caprellidae was represented in nine of the transect stations and one selected station with a high count of 25 individuals at station 11 (average 6 ± 2). Unidentified amphipods, Ampithoe sp., and Corophium sp. were also present in densities of 21 per station or less. Other groups present included nematodes, nemerteans, ectoprocts, oligochaetes, tanaids, sea algae fragments, and anemones.

A correlation between total wet weight of eelgrass and number of species present was tested for the combined sample (four selected samples and 10 transect samples) employing a regression analysis program. The correlation between biomass of eelgrass and number of species was highly significant (Table 5). The correlation between biomass and species was also evident by comparing the number of species in samples from areas of low eelgrass cover (selected samples) with the numbers of species from areas with higher eelgrass cover (transect samples). The average number of species in the selected samples was 4 ± 3 compared with 22 ± 2 for the transect samples. The average wet-weight eelgrass biomass of the selected samples was 8.5 ± 1.2 g compared with 164 ± 40 g for the transect stations.

1.8 Operl Island and Cape Glazenap

Three similar sand-spit beaches were sampled near the west entrance to Izembek Lagoon, Alaska Peninsula (Figs. 1, 9, and 16; Table 1).

The Operl Island site (Fig. 14) was on the seaward side of Operl Island, a narrow barrier sand spit about 16 km in length. Two cores were collected, one at low tide at the water's edge and the other in dry sand at about mid-tide level. The beach was exposed to the full force of the ocean surf and was composed of loose, coarse, volcanic sand. No macrofauna were found but numerous razor clam (*Siliqua patula*) shells littered the beach at all tide levels, evidence of a subtidal population of clams offshore. Scattered irregularly in the mid- to high-tidal zone were distinctive patches of dark dry sand, ranging in size up to about 100 meters in area (Fig. 15). Later analysis revealed that these patches were of sand grains which were magnetically attracted to each other, causing their striking appearance compared with adjacent unmagnetized sand. This was the only site in the Bering Sea and Norton Sound where magnetic sand was observed. No organisms were found in later examination of the two core samples from Operl Island.

Table 5. Analysis of variance of linear regression for number of species vs. wet weight of eelgrass at Izembek Lagoon.

Source	DF	SS	MS	F
Total	13	914		
Regression	1	678.8	678.8	34.6**
Residual	12	235.2	19.6	

R-Square = 0.74

DF = Degrees of freedom

SS = Sums of squares

MS = Mean squares

F = F-distribution value

** = Significant at the 1% level of testing

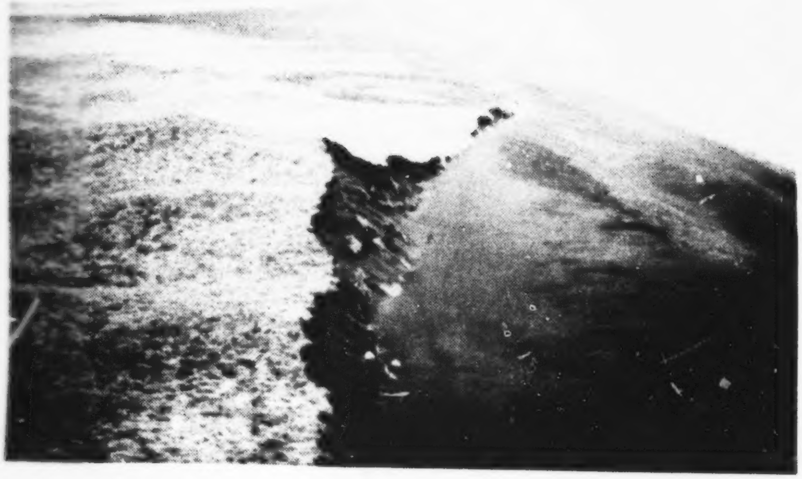

Fig. 14. Operl Island sand sample site. Bering Sea on right. 16 June 1976.

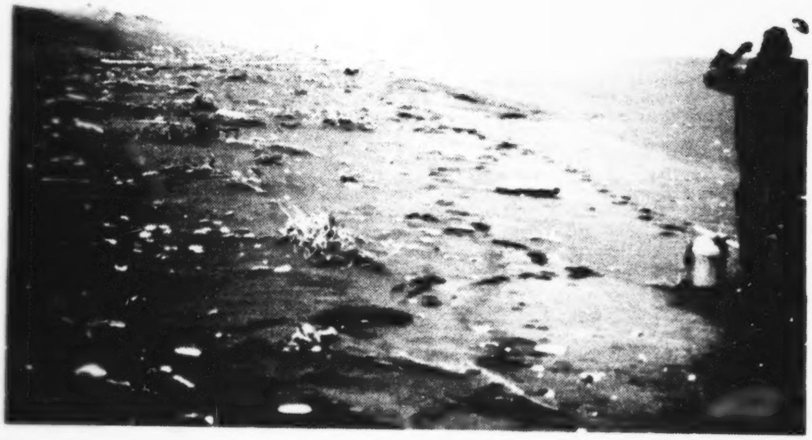

Fig. 15. Operl Island site. Light-colored patch about 2 m in diamater to left of figure is black magnetic sand. Small, light-colored objects are razor clam shells and pumice rocks. Bering Sea on right; grass covered dunes on left. 16 June 1976.

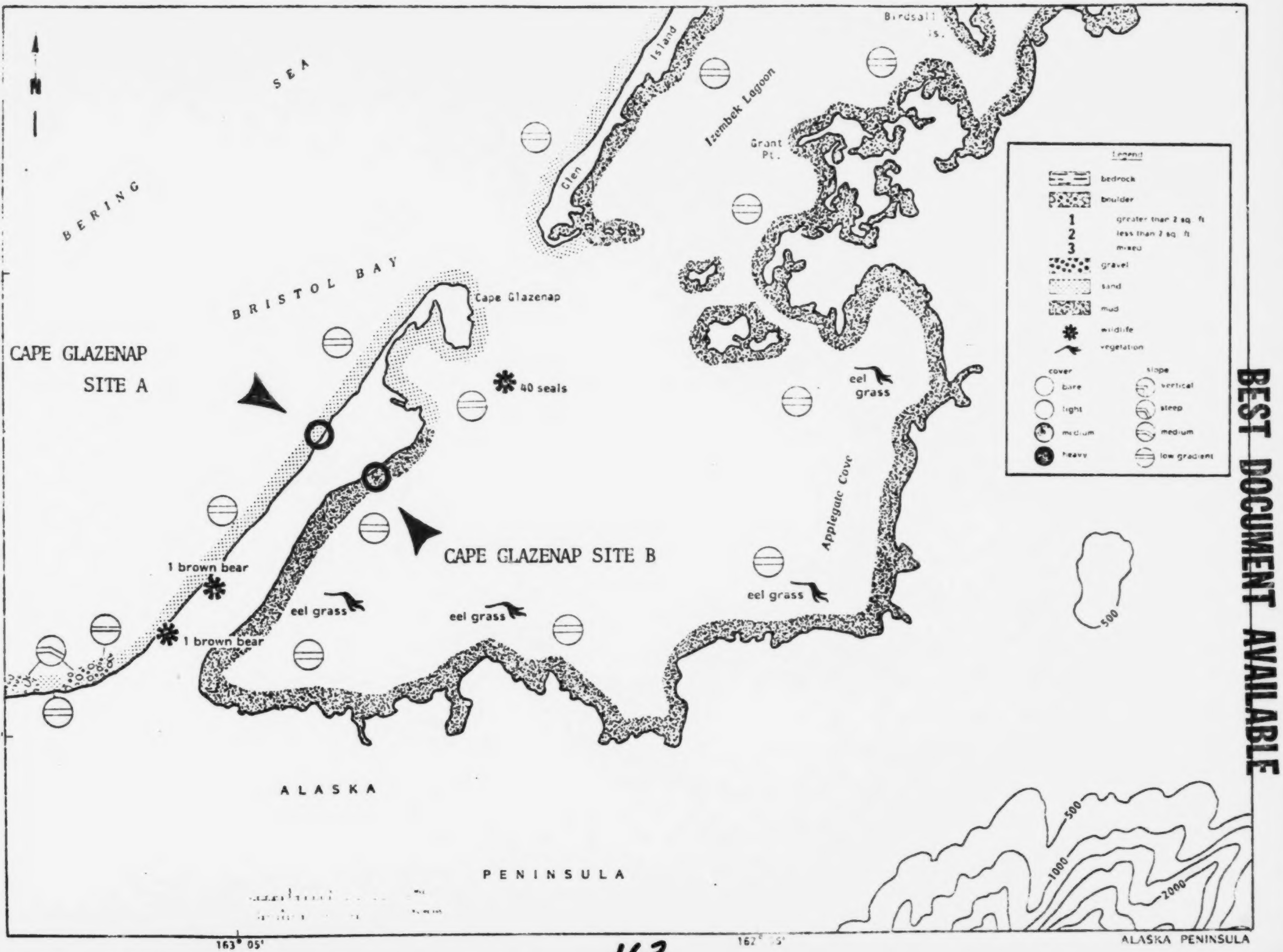
N
BERING SEA
BRISTOL BAY
CAPE GLAZENAP SITE A
CAPE GLAZENAP SITE B
Cape Glazenap
Glen Island
Izembek Lagoon
Grant Pt.
Birdsall Is.
Applegate Cove
40 seals
1 brown bear
1 brown bear
eel grass
eel grass
eel grass
eel grass
ALASKA
PENINSULA
55° 15'
55° 10'
163° 05'
162° 55'
500
500
1000
2000
Legend
bedrock
boulder
1 greater than 2 sq. ft
2 less than 2 sq. ft
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
ALASKA PENINSULA

Fig. 16. Location of sites at Cape Glazenap.

The Cape Glazenap site (Figs. 1 and 16) was near the eastern extremity of Cape Glazenap. Four cores were collected, two on the seaward side of the Cape (Site A) and two on the lagoon side of the Cape (Site B). At each of the two sites, as at Operl Island, one core was collected at low tide at the water's edge and another was collected at about the mid-tide level.

The 1-liter core at the water's edge on the seaward side of Cape Glazenap contained 73 polychaetes of a single genus, Saccocirrus, which was unexpected on this apparently unproductive high-energy beach. The other core sample from the mid-tide level had only a few fragments of Fucus sp., sea grass (probably Zostera marina), and polychaetes. The site is adjacent to biologically-productive Izembek Lagoon, so most biotic fragments probably originated in the lagoon.

The two core samples from the lagoon side of Cape Glazenap contained no recognizable biota, despite this site's location inside the lagoon. The lack of organisms was probably attributable to the unstable and constantly shifting silt-sand substrate.

1.9 Amak Island

Amak Island is a rocky island of volcanic origin located in Bristol Bay northwest of Izembek Lagoon on the Alaska Peninsula (Figs. 1 and 17; Table 1). The shoreline has stretches of beach boulders broken by steep to vertical cliffs. The area we sampled is on the southwest shore of Amak Island, an area of low bluffs flanking a bedrock and medium-boulder beach (Fig. 18).

The intertidal study site on Amak Island yielded 101 species of algae and invertebrates. The most conspicuous features along our 75 m transect were a band of Fucus sp. from 1 m through 49 m, and a band of Alaria spp. from 25 m through 68 m; Alaria provided greater biomass than did Fucus. Many of

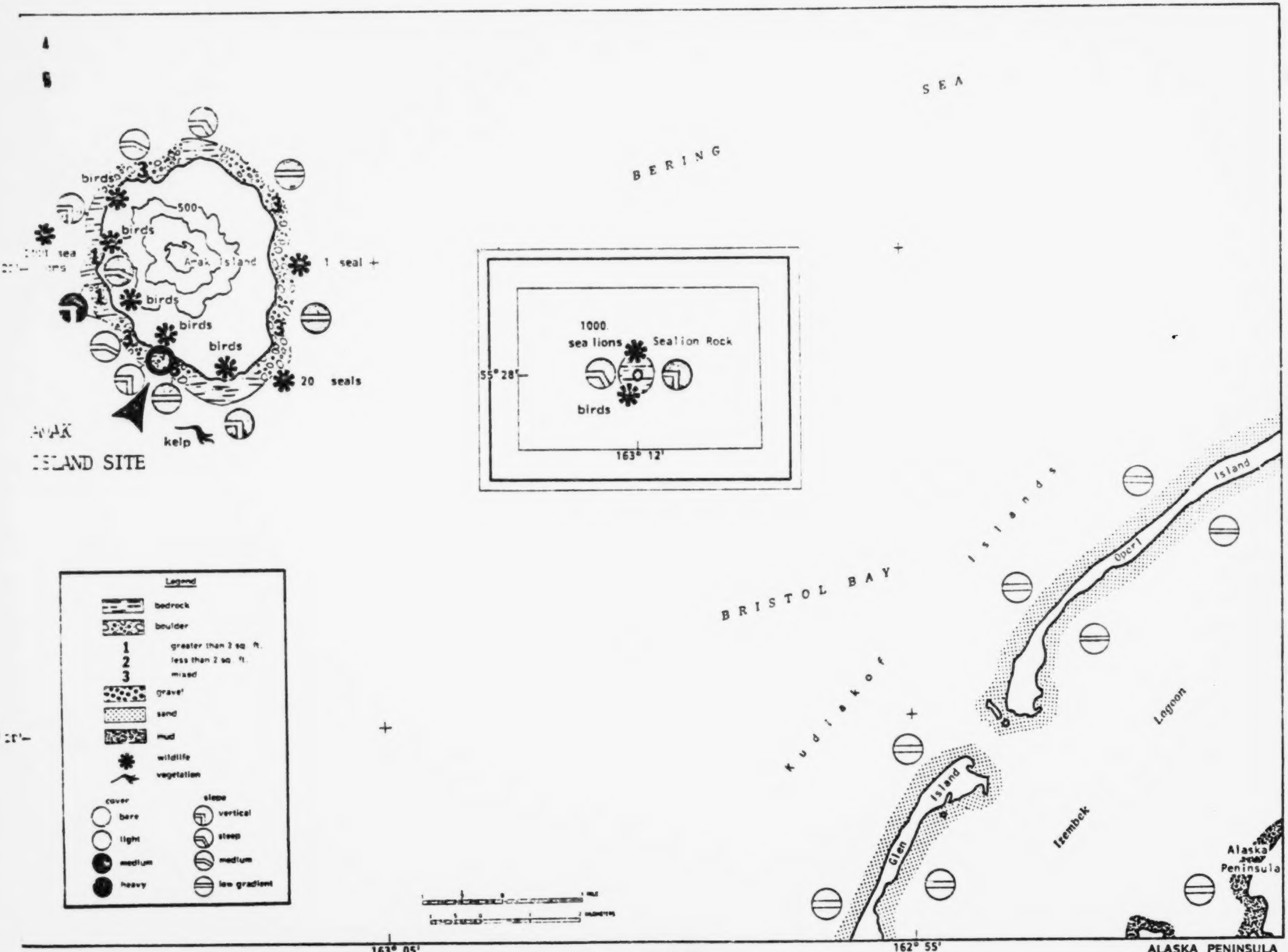
SEA
BERING
BRISTOL BAY
Kudiakof Islands
Operl Island
Glen Island
Izembek Lagoon
Alaska Peninsula
ALASKA PENINSULA
163° 05'
162° 55'
55° 28'
163° 12'
1000 sea lions
Sealion Rock
birds
Amak Island
500
birds
1 seal
20 seals
kelp
AMAK ISLAND SITE
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 17. Location of site at Amak Island.

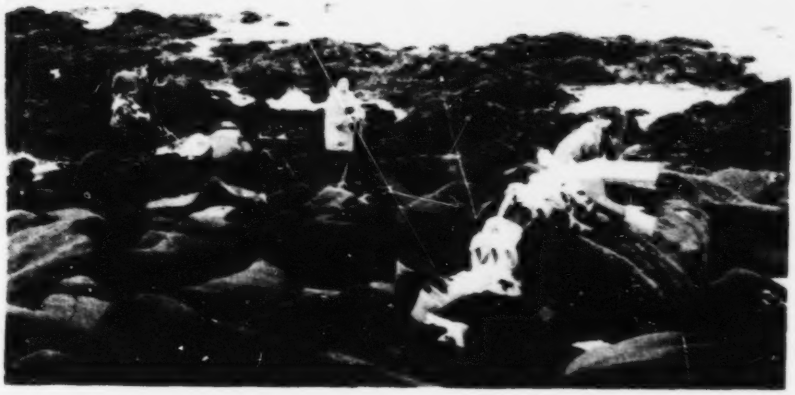

Fig. 18. View of transect at Amak Island sampling site. 19 July 1975.

the medium boulders in the low zone typically had a heavy cover of Alaria sp., coralline algae, and sponges. The invertebrates were predominantly smaller forms such as polychaetes, small gastropods, nestling pelecypods, amphipods, and isopods. Mussels were sparse. We did not find any Mytilus, but Modiolus modiolus was collected in two quadrats. The barnacles Balanus glandula, B. cariosus, and Chthamalus dalli were collected in small numbers from a total of nine quadrats for the three species. Few predators were observed. Leptasterias hexactis and Ophiopholis aculeata were the only sea stars collected and were not abundant. The carnivorous snails Nucella lima and N. lamellosa were each collected in two quadrats. Although there were bird rookeries in the area, no birds were observed in the immediate vicinity of our study area, and there were no obvious signs of predation by birds.

1.10 Cape Lapin

The Cape Lapin site, on the north side of Unimak Island, lies below a high bluff headland facing north (Figs. 1 and 19; Table 1). The substrate of the site consists of large and small boulders and rubble pavement. To either side of the sample area are sandy beaches flanked by eroding banks. Further to the east and west, high bluffs predominate. Exposure of the site to surf and current-transported sand and sediment was evident.

Four transects were established: one running at 340° magnetic and at right angles to the shoreline and three placed horizontally at right angles to the normal transect. Sampling on the normal transect was done systematically with a 1/16 m^2 quadrat every 5 m, beginning at 0 m, to 40 m, which was the level of low tide on 17 June 1976. The three parallel transects were established by a stratifying procedure at the 8, 19, and 32 m levels of the normal transect. The 8 m transect fell in a relatively bare zone occupied primarily by Littorina sitkana and Balanus glandula. The 19 m transect fell

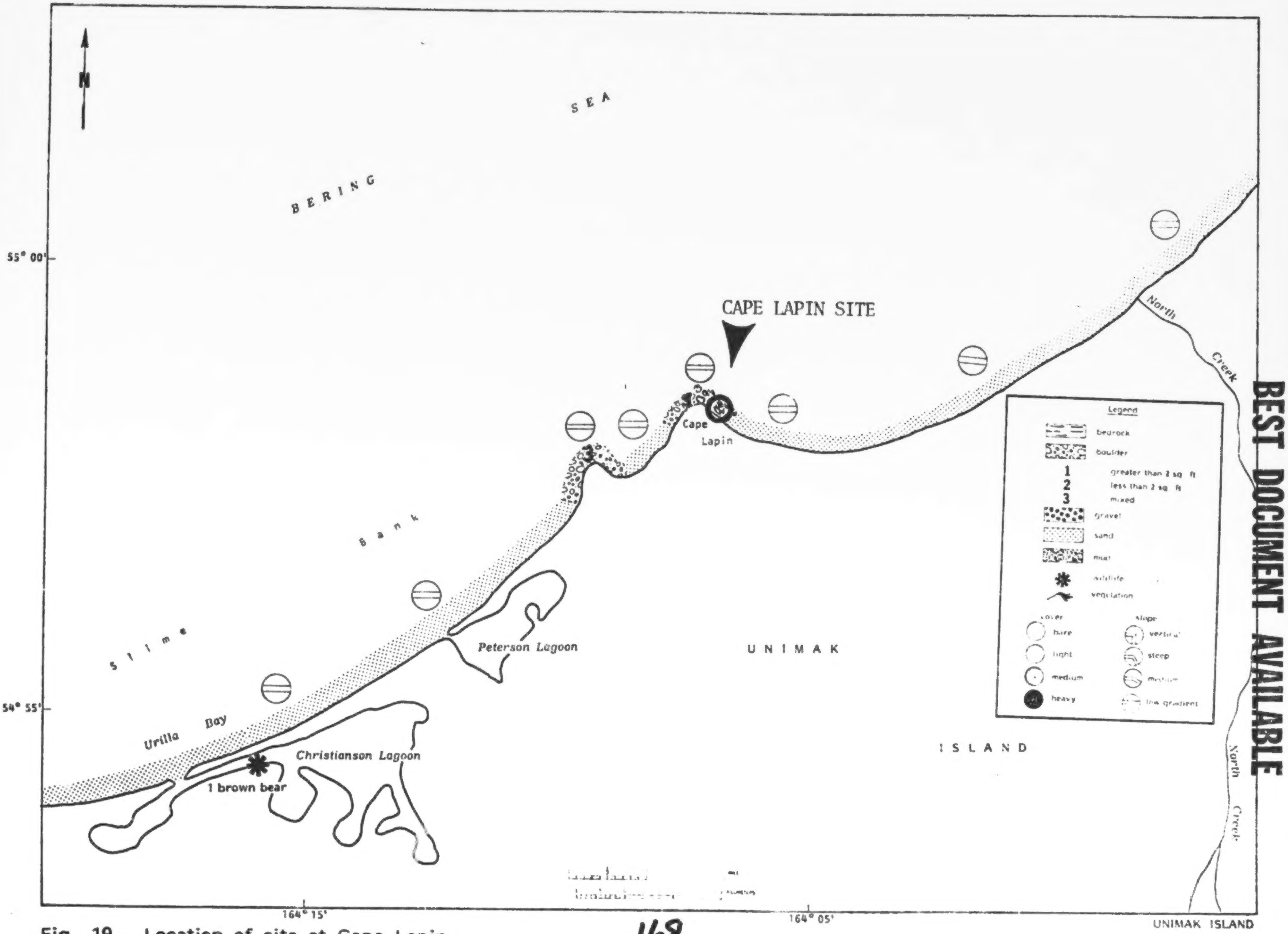
N
BERING SEA
CAPE LAPIN SITE
Cape Lapin
Bank
Slime
Peterson Lagoon
UNIMAK
ISLAND
Urilla Bay
Christianson Lagoon
1 brown bear
North Creek
North Creek
55° 00'
54° 55'
164° 15'
164° 05'
UNIMAK ISLAND
Legend
bedrock
boulder
1 greater than 2 sq ft
2 less than 2 sq ft
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 19. Location of site at Cape Lapin.

in a zone dominated by *Mytilus edulis* and *Balanus*. The 32 m transect fell in a zone dominated by *Mytilus*, *Balanus*, and *Fucus distichus*.

Sampling in the horizontal transects followed a randomized nested sampling design. Each transect was subdivided into 50 contiguous 1 m plots (referred to as plots in the rest of this section), and two plots along each transect were selected randomly. Each plot was subdivided into 1/16 m^2 quadrats of which two were sampled randomly. Random sampling within plots permitted averaging and variance calculations and the application of normal sampling theory for hypothesis testing. A further analysis of the Cape Lapin data will be included in a separate manuscript to be published later.

The biota was subdivided into three categories depending upon relative wet-weight biomass of the species or group. A dominant-biomass category included each species or group that was represented in one or more samples with a biomass exceeding 400 g wet weight. A secondary-biomass category included species and groups in which at least one or more samples contained a species less than 80 g wet weight and greater than 0.3 g wet weight. A small-sized category, designated the tertiary biomass category, contained species and groups of 0.8 g wet weight or less. The highest wet weight of any species or group established the biomass category for all other samples belonging to the same species or group.

To facilitate graphical descriptions of the biota, the species and groups were assigned reference numbers (Table 7).

The distribution of total biomass in random pairs of quadrats within plots by dominant, secondary, and tertiary categories is shown in Table 6. Biomass was lowest in the *Littorina sitkana*-*Balanus glandula* stratum (LB) within the elevation range of 2.1 to 2.5 m when compared to the *Mytilus edulis*-*Balanus* (MB) and the *Mytilus*-*Balanus*-*Fucus distichus* (MBF) strata at the 1.0 to 1.3 m and the 0.8 to 1.1 m elevations, respectively.

Table 6. Total biomass (g) and number of species or groups (in parentheses) in pairs of 1/16 m^2 quadrats taken from m^2 plots in the *Littorina sitkana*-*Balanus glandula* (LB) stratum, *Mytilus edulis*-*Balanus* (MB) stratum, and the *Mytilus-Balanus*-*Fucus distichus* (MBF) stratum, Cape Lapin.

Strata	Unit	Dominant	Secondary	Tertiary	Total
LB	meter 1	9.95 (2)	0[a]	0	9.95 (2)
	meter 2	2.90 (3)	0[a]	0	2.90 (2)
MB	meter 1	599.7 (1)	160.56 (2)	0.84 (15)	761.10 (2)
	meter 2	111.2 (1)	29.9 (5)	0.03 (5)	141.13 (11)
MBF	meter 1	93.1 (2)	4.3 (6)	0.32 (7)	97.72 (15)
	meter 2	729.4 (2)	126.7 (6)	2.90 (19)	859.00 (27)

a. Secondary biomass for the LB stratum is classified as dominant biomass.

Table 7. Key to the species and groups identified in Cape Lapin samples.

1A. *Fucus distichus*
1B. *Bangia fusco-purpurea*
1C. *Monostroma* sp.
1D. *Desmarestia* sp.
1E. *Porphyra* sp.
1F. *Pterosiphonia bipinnata*
1G. *Odonthalia* cf. *washingtoniensis*
2. *Mytilus edulis* (>2.0 cm)
3. *Mytilus edulis* (1.5-2.0 cm)
4. *Mytilus edulis* (<1.5 cm)
5. *Balanus glandula*
6. *Collisella pelta*
7. *Littorina sitkana*
8. *Nucella lima*
9. brown algae fragments
10. red algae fragments
11. flatworms
12. nemerteans and fragment(s)
13. *Emplectonema gracile*
14. nematode(s)
15. *Nereis vexillosa*
16. scale-worm fragment(s)
17. *Pholoe minuta*
18. Phyllodocidae
19. *Eteone longa*
20. *Typosyllis a. admantea* (epitoke condition not specified)
21. *Typosyllis a. admantea* with epitoke
22. *Typosyllis a. admantea*
23. *Sphaerodoropsis minuta*
24. *Sphaerodoropsis sphaerulifer*
25. Spionidae
26. *Polydora quadrilobata*
27. *Fabricia sabella*
28. *Fabricia crenicollis*
29. Oligochaete
30. Gastropod
31. *Barleeia* sp.
32. *Pelecypod*
33. *Turtonia occidentalis*
34. *Idotea wosnesenskii*
35. *Paramoera columbiana*
36. Diptera larvae
37. sea anemone
37A. hydroid
38. sea cucumber
38A. *Cucumaria pseudocurata*
39. *Ischyrocerus anguipes*

The distribution of mean biomass in a quadrat averaged over pairs of quadrats within plots, and numbers of species and groups in dominant, secondary, and tertiary categories is shown in Figure 20. Depending upon the stratum and plot, the dominant biomass category contained between 1 and 2 species, followed by secondary biomass composed of 5 or 6 species and groups, and the tertiary biomass composed of 5 to 20 small species and groups (Table 7, Figure 20).

The second plot of the MBF stratum (Table 6) was richest in numbers of species and contained several more tertiary species and groups with higher biomass than other plots of the MBF stratum and the plots of the LB stratum.

The number of individuals of species in the dominant and secondary biomass categories is presented in Table 8. Balanus glandula was numerically most abundant, followed by Littorina sitkana and small (<1.5 cm) Mytilus edulis. Species common to every stratum were Balanus glandula, Collisella pelta, and Littorina sitkana.

For the dominant and secondary biomass categories, the least number of individuals occurred in the LB stratum (Table 6). Variation in numbers of individuals was pronounced for all size categories of Mytilus edulis when compared to other species (Table 8). Within plots, Balanus glandula showed the highest variability.

For only the large and small size categories of Mytilus (see Table 8), pairs of 1/16 m^2 quadrat samples were more consistently similar in numbers of individuals when comparisons were made within plots than between plots in the same stratum. Pairs of quadrat samples within plots for small Mytilus edulis, Collisella pelta, and Littorina sitkana were slightly less similar; Balanus glandula samples were least similar. For quadrat pairs within plots, the number of individuals of Nucella lima and Fucus distichus were too small to compare.

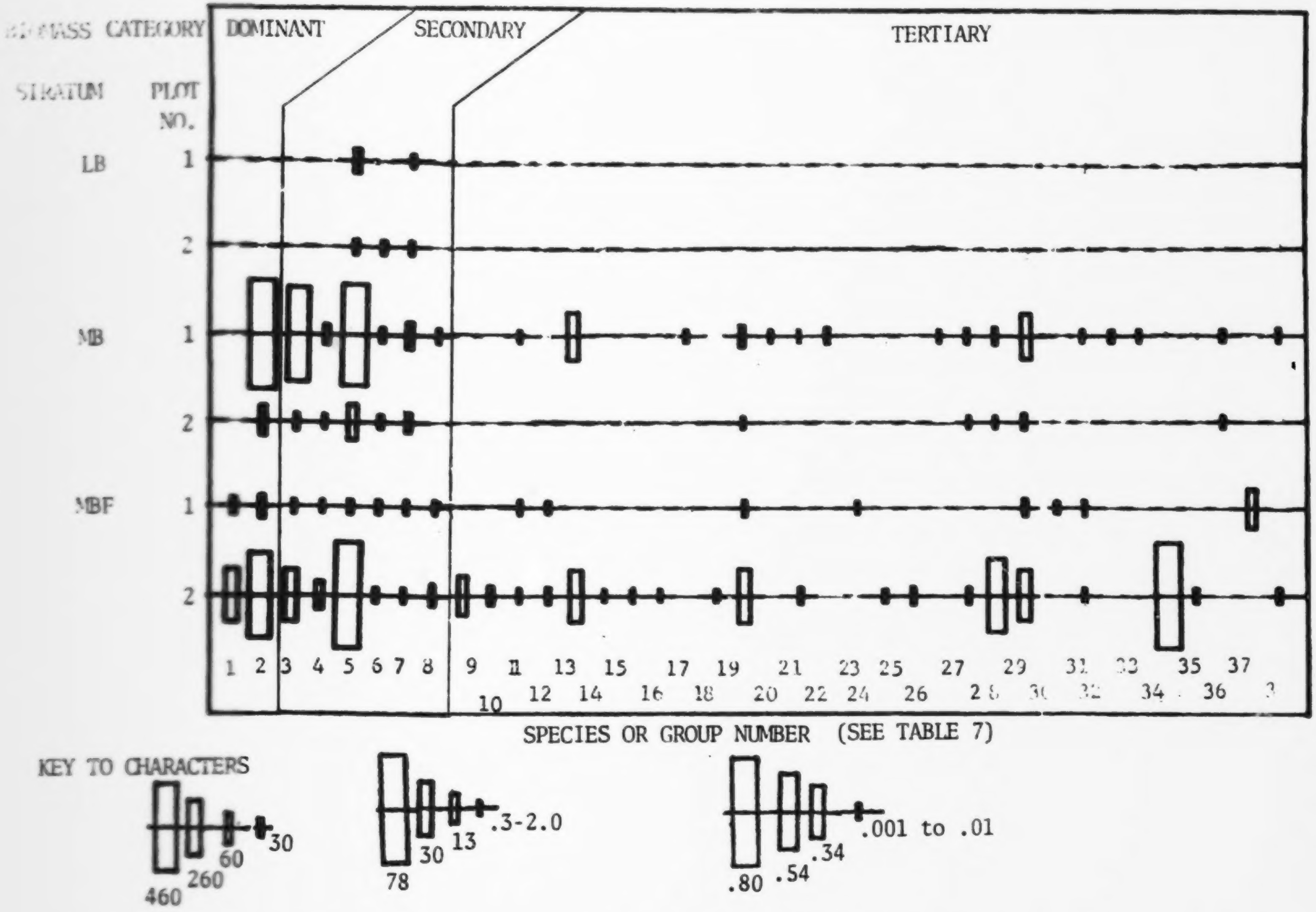

Fig. 20. Mean biomass per 1/16 m^2 quadrat in randomized meters within lateral transects at Cape Lapin, Unimak Island, Alaska. LB = *Littorina sitkana* - *Balanus glandula* stratum; MB = *Mytilus edulis* - *Balanus* stratum; MBF = *Mytilus-Balanus-Fucus distichus* stratum. Characters shown in the key represent mean biomass (in grams) in dominant, secondary, and tertiary categories, respectively.

Table 8. Numbers of individuals of species in the dominant and secondary biomass categories by stratum, Cape Lapin. Data from two 1/16-m^2 quadrats in each plot.

	Stratum											
	LB				MB				MBF			
	meter				meter				meter			
Species	1		2		1		2		1		2	
Fucus distichus					0	0	0	0	1	0	5	0
Mytilus edulis (> 2.0 cm)					52	85	21	7	78	85	5	6
Mytilus edulis (1.5-2.0 cm)					13	20	3	8	39	41	0	2
Mytilus edulis (< 1.5 cm)					15	38	1	6	113	47	0	2
Balanus glandula	110	29	11	10	94	255	90	4	121	2	0	167
Collisella pelta			1	0	40	37	7	11	92	15	18	13
Littorina sitkana	15	8	3	7	70	133	15	57	36	9	16	37
Nucella lima					2	0	0	0	0	1	0	1

LB = *Littorina sitkana*-*Balanus glandula* stratum

MB = *Mytilus edulis*-*Balanus* stratum

MBF = *Mytilus*-*Balanus*-*Fucus distichus* stratum

Analysis of the total number of individuals in sample quadrats for the tertiary biomass category showed oligochaetes to be the most common organisms present. They appeared in all samples taken, both 1/16 m^2 quadrats and m^2 plots. *Eteone longa* also occurred frequently, appearing in every plot but not in every quadrat. *Fabricia crenicollis* was the most abundant species with 1416 individuals observed in a single quadrat; Oligochaeta was only slightly less abundant. *F. crenicollis* and *F. sabella* were common to both lower-elevation strata but were not present in every plot.

Analysis of the distribution of biomass along 5-m intervals of a transect normal to the shoreline for the systematic sample showed *Balanus glandula* and *Mytilus edulis* to be the most common species. These organisms contributed the dominant biomass at the 15, 20, 25, and 35 m sites.

Secondary biomass of the systematic sample contained six species (four animal and two plant) of which *Littorina sitkana* was most common, followed by *Collisella pelta*. *Littorina* contributed the highest biomass for sites at 5, 10, and 30 m. The algae *Bangia fusco-purpurea* and *Monostroma* sp. contributed the highest secondary biomass at the lowest elevation site, 40 m.

Tertiary biomass of the systematic sample contained 21 species or groups of which *Odonthalia* cf. *washingtoniensis* contributed the highest biomass. The most common species were oligochaetes which occurred in four of the nine systematic samples.

1.11 Cape Mordvinof

Cape Mordvinof is on the northwest shore of Unimak Island, along the southwestern boundary of Bristol Bay (Figs. 1 and 21; Table 1). The substrate at the Cape is varied. Adjacent to our study area is a black sand beach cut by a stream and a lowlying area of boulders and tidepools where *Fucus distichus* and *Odonthalia floccosa*, both covered with the epiphyte

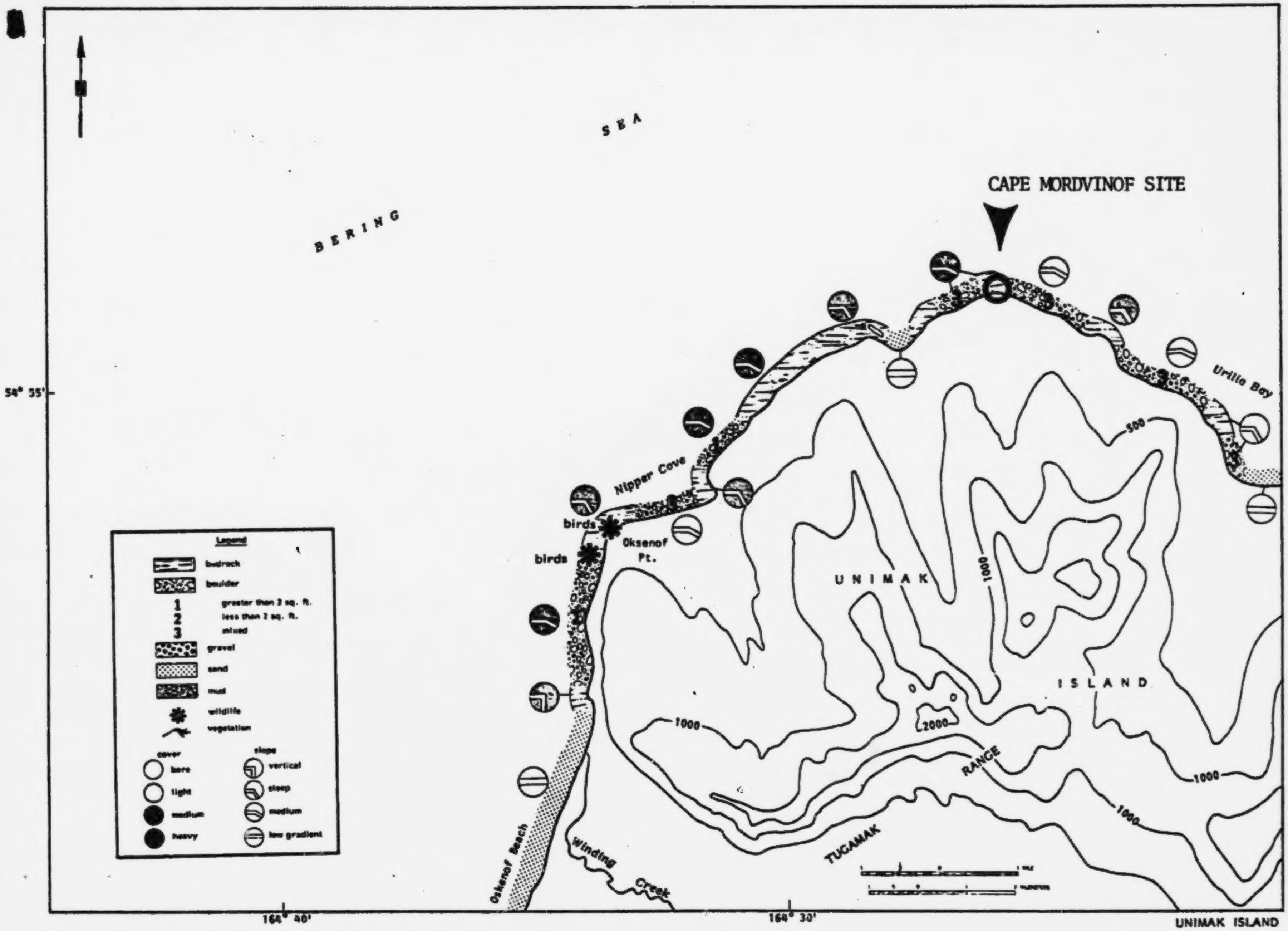
N
BERING
SEA
CAPE MORDVINOF SITE
Urilia Bay
Nipper Cove
birds
birds
Oksenof Pt.
Oksenof Beach
Winding Creek
UNIMAK
ISLAND
TUGAMAK
RANGE
500
1000
1000
1000
1000
1000
2000
54° 55'
164° 40'
164° 30'
UNIMAK ISLAND
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 21. Location of the site at Cape Mordvinof.

176

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Soranthera ulvoides, were abundant along with large patches of filimentous green algae. The most conspicuous invertebrates in this peripheral area were Balanus balanoides, B. cariosus, and Mytilus edulis.

We established our transect on a nearby rocky point and an additional zonation-study site on a large isolated boulder (Fig. 22). This boulder had an almost vertical, smooth face and dramatically exhibited distinct horizontal banding--but proved rather depauperate. The highest zone was dominated by the red alga Halosaccion glandiforme along with oligochaetes and the herbivorous gastropod Littorina sitkana. The next band was characterized by the presence of the red algae Porphyra sp. and Callophyllis flabellulata and the isopod Idotea wosnesenskii. Because of tidal and surf conditions, no samples were taken from the two lowest zones where Alaria sp. and an unidentified bladed red alga predominated.

The bedrock substrate along the transect had a shelf-like appearance; the substrate formed a series of plateaus separated by rather abrupt breaks. The highest station on the transect (M19) occurred in a tidepool at 2.77 m elevation and was dominated by Odonthalia floccosa, which occurs frequently in low-gradient wet areas. The nestling bivalve Turtonia occidentalis, often found with Odonthalia, was abundant as were the gastropods Littorina sitkana and Lacuna marmorata. Mytilus edulis and limpets were also present. A second station (M15) occurred at the same elevation but was not in a tidepool. Here the predominant invertebrate was Balanus glandula; a moderate number of littorines and small amounts of the red algae Porphyra sp. and Halosaccion glandiforme were also collected. Two quadrats (M12 and M9) occurred at approximately the same level in the mid-tidal zone, on a plateau at 2.1 m elevation. These samples contained primarily Porphyra sp. and the barnacles Balanus glandula and Chthamalus dalli. Of the two barnacles, C. dalli was more numerous at this station.

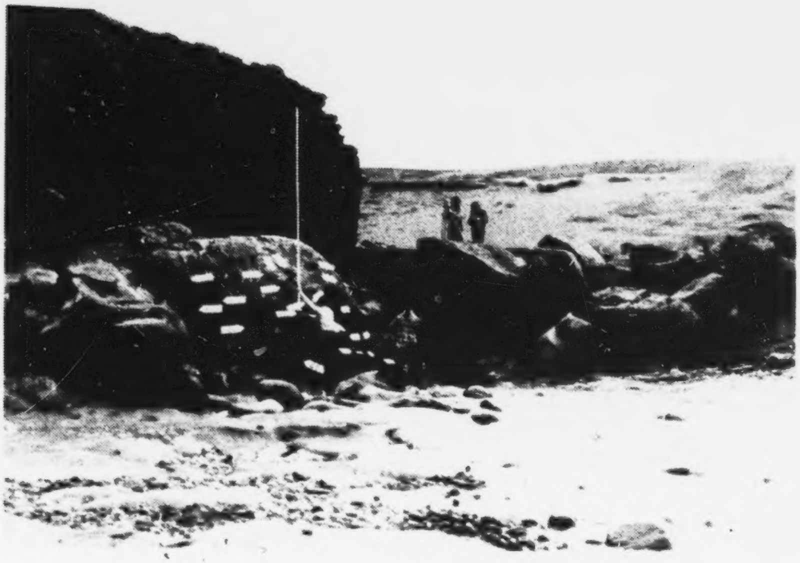

Fig. 22. Cape Mordvinof sampling site. 24 July 1975.

Two stations (M6 and M0) located in the lower intertidal zone exhibited the greatest species richness at the Cape Mordvinof site; at both, *Balanus cariosus* contributed the greatest biomass per sample. The heavy plates of *B. cariosus* and the interstices between individual barnacles provide shelter and protection for numerous small organisms.

At station M6 (elevation 0.98 m), *Mytilus edulis* contributed the second greatest biomass and *B. glandula* third in abundance. *Turtonia occidentalis*, nestling on the red alga *Pterosiphonia bipinnata*, was also found at this station as were limpets, sea anemones, *Cucumaria curata*, *Littorina aleutica*, two kinds of oligochaetes, several polychaetes, and coralline algae. At station M0 (elevation 0.30 m), subordinate species included *Katharina tunicata*, *Leptasterias hexactis*, *Collisella pelta*, *Chthamalus dalli*, and *Alaria praelonga* (represented by one large individual, 51.8 g wet weight). *Turtonia occidentalis* and the clam *Hiatella arctica*, which attaches with byssal threads or bores into the substrate, were both collected. These organisms were probably associated with the *Alaria* holdfast. Nine species of polychaetes occurred in this quadrat, most of which probably found shelter in the kelp holdfast or in the interstices of barnacles.

A third station (M3) in the low intertidal zone at an elevation of 0.78 m was dominated by *Balanus glandula*. We collected 374 large *B. glandula* (average size, approximately 20 mm) in this quadrat and noted that 90% of the quadrat was covered with newly-settled juvenile barnacles. Small limpets were also abundant at this station.

1.12 Sennet Point, Unimak Island

Sennet Point is south of Cape Sarichef at the southwestern end of Unimak Island (Figs. 1 and 23). It is exposed to the full force of storms from both the Bering Sea and the Gulf of Alaska, although the sea was calm

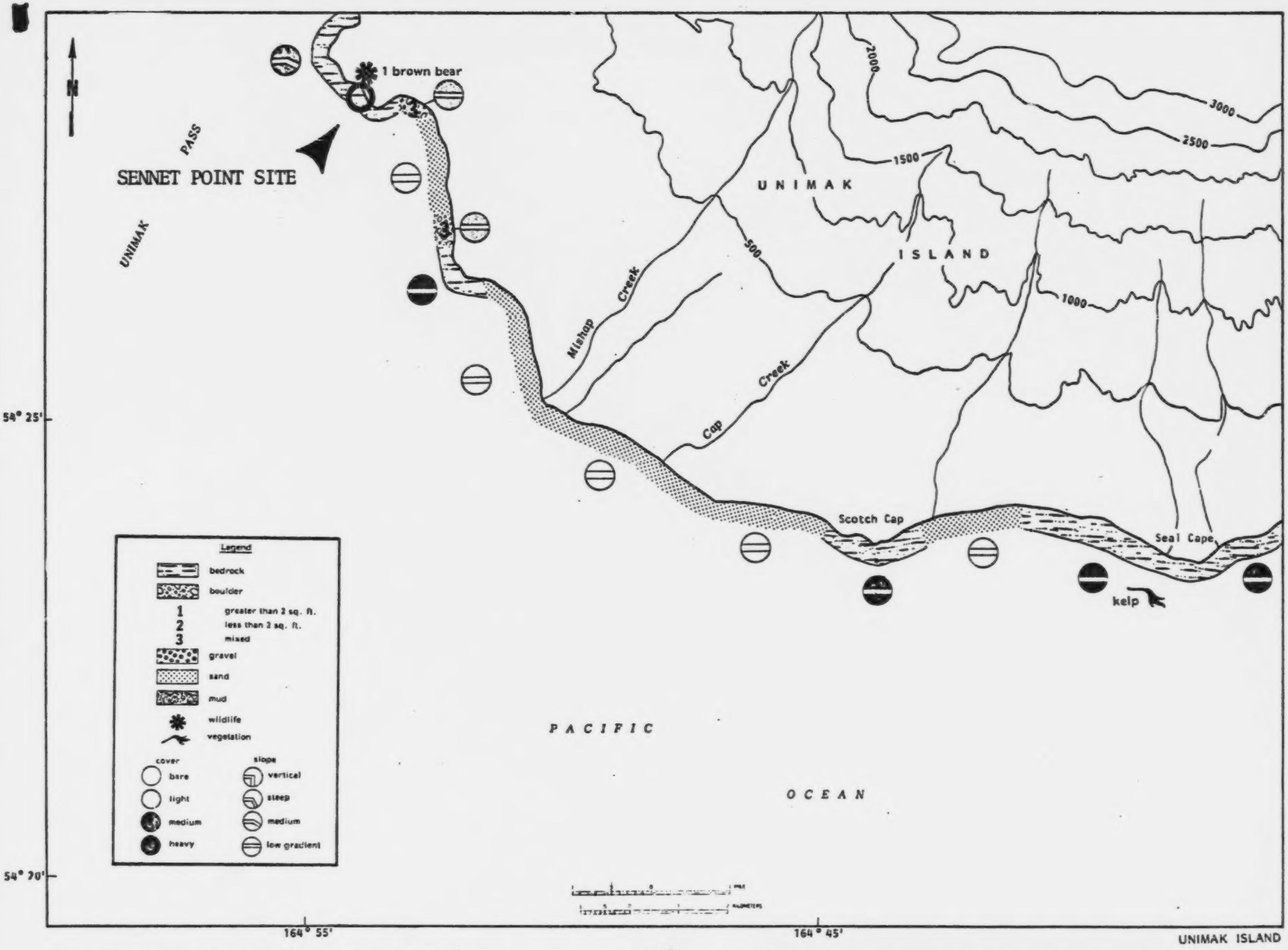
1 brown bear
N
PASS
SENNET POINT SITE
UNIMAK
UNIMAK
ISLAND
2000
3000
2500
1500
500
1000
Mishap Creek
Cap Creek
Scotch Cap
Seal Cape
kelp
54° 25'
54° 20'
164° 55'
164° 45'
PACIFIC
OCEAN
UNIMAK ISLAND
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 23. Location of site at Sennet Point.

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during our sampling. We sampled a bedrock bench with boulders and rubble on its surface (Fig. 24; Table 1). One quadrat (containing no macroscopic organisms) was located above Mean Higher High Water (MHHW). The remaining six samples were collected between MHHW and mean lower low water. Four of the six samples containing organisms were taken from the upper intertidal zone (MHHW to mean tidal level). We excluded the plot containing no organisms from the following analysis.

Littorina sitkana was the only species found in all samples; it had the highest average rank in wet weight (Table 9). Other important herbivores which occurred in the samples were *Collisella* spp. and *Katharina tunicata*. The average ranks of wet weight of *C. pelta* and *K. tunicata* were high (Table 9), but the other herbivores *Collisella* sp., *C. digitalis*, and *C. strigatella* occurred infrequently (frequency of occurrence, f $\leq$0.33) and were low in abundance ($\leq$10 individuals per sample) and small (wet weight $\leq$0.2 g).

A single individual of *Nucella lima* occurred in one sample. *Nucella* was the only major predator (one which could play a major role in community dynamics) in our samples. This is not surprising because quadrat size and the number of samples taken were too small to adequately estimate the population sizes of major predators.

Among the primary competitors for space at Sennet Point, *Mytilus edulis* was uncommon (found in two quadrats; abundance, 2 and 19), and individuals were small (shell length $\leq$ 1.5 cm) in our samples. *Balanus cariosus* (BC), *B. glandula* (BG), and *Chthamalus dalli* (CD) were relatively uncommon (each species occurred in 33% of our samples) and were highly variable in abundance (range of number of individuals per 1/16 m^2 was: BC, 4-33; BG, 47-223; CD, 65-1154).

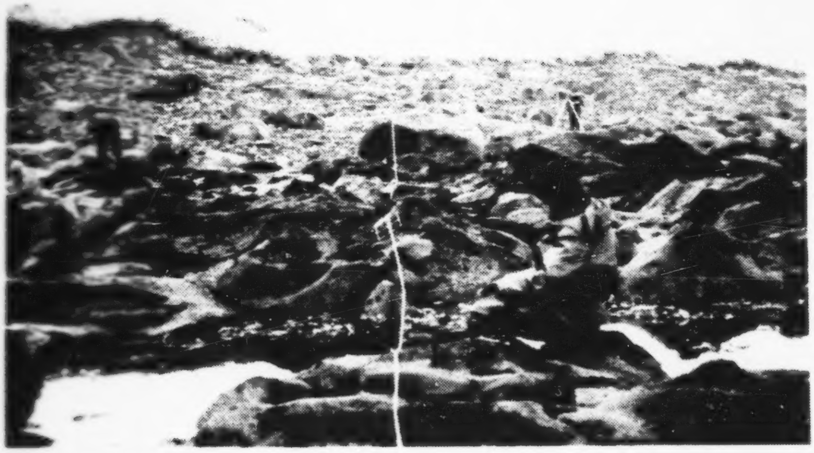

Fig. 24. Sennet Point site; view of upper portion of Transect 2. 8 June 1976.

Table 9. Statistics for the 10 species of invertebrates showing the greatest average rank on the transect at Sennet Point, Unimak Island.

Species	Average rank[a]	Wet weight(g)[b]	Frequency of occurrence[c]	Abundance[d]	Dominance[c]
Littorina sitkana	13.7	3.4, 0.002-51.2	1	23, 1-391	1/6
Porphyra sp.	10.7	32.2, 0.06-35.1	0.5	--	2/6
Collisella pelta	10.3	~3.4, ~1.3-4.4	0.5	18, ~9-19	0
Katharina tunicata	10.2	15.7, 8.4-23	0.33	1	1/6
Alaria sp.	10.2	10.7, 2.4-19	0.33	--	0
Corallinaceae (crustose)	9.2	3.3, 1.4-5.2	0.33	--	1/6
Cucumaria pseudocurata	8.5	0.7, 0.3-1.1	0.33	34, 22-47	0
Idotea fewkesi	8.3	0.1, 0.009-0.2	0.5	3, 3-4	0
Halosaccion sp.	8.2	0.4, 0.1-0.8	0.33	--	0
Tonicella lineata	8.0	1.8, 1.7-1.9	0.33	1, 1-2	0

a. Rank of wet weight of each species averaged over 6 quadrats (1, lowest weight). Species listed in order of decreasing rank.

b. Median and range of wet weights per liter in all quadrats where the species was found.

c. Proportion of quadrats that contained the species.

d. Animals only; median and range of number of individuals per 1/16 m^2 in all quadrats where the species was found.

e. Proportion of quadrats in which the species was among those making up 50% of the wet weight. Summation in each quadrat began with the heaviest species.

1.13 Akun Island

Akun Island is in the Aleutian Islands and is the northernmost island of the Krenitzin group. Akun Island lies 23 miles southwest of Unimak Island (Figs. 1 and 25; Table 1). Our study site was on the southwest shore of Akun Island, facing Akun Strait, and was somewhat protected from ocean waves by offshore rocks and reefs. The waters in the vicinity of Akun Island are well known for rip tides and strong currents. In Akun Strait, between Akun and Akutan Islands, currents have been estimated at a maximum velocity of 12 knots. Fig. 26 shows the study site at slack water.

The reef we sampled had a heavy cover of algae and invertebrates which was composed of a high number of species. We collected 156 species on Akun Island as compared to 101 species on Amak Island. Three species of barnacles were found (*Balanus glandula*, *B. cariosus*, and *Chthamalus dalli*); *Balanus cariosus* was most abundant, being found in 18 of 20 quadrats. The mussels *Mytilus edulis* and *Modiolus modiolus* were each found in two quadrats. Only small (<1.5 cm) *Mytilus* were present in one quadrat, and in the second, which contained mainly large (>2.0 cm) *Mytilus*, 18 of 21 individuals were dead. We found 34 species of polychaetes in samples collected along the 58 m transect; no species occurred in more than 11 of 20 quadrats and several species occurred in only one quadrat. Gastropods were especially abundant and well distributed. We found 13 species of gastropods of which *Littorina sitkana*, *Margarites helicinus*, and *Barleeia* sp. all occurred in 13 or more quadrats. The nestling bivalves *Turtonia occidentalis* and *Musculus discors* occurred in than half the quadrats and exhibited the greatest number per quadrat of any species. *Turtonia occidentalis* averaged 1539 individuals per quadrat. Predators were not abundant. The carnivorous snail *Nucella lima* was found in seven quadrats, and the small sea

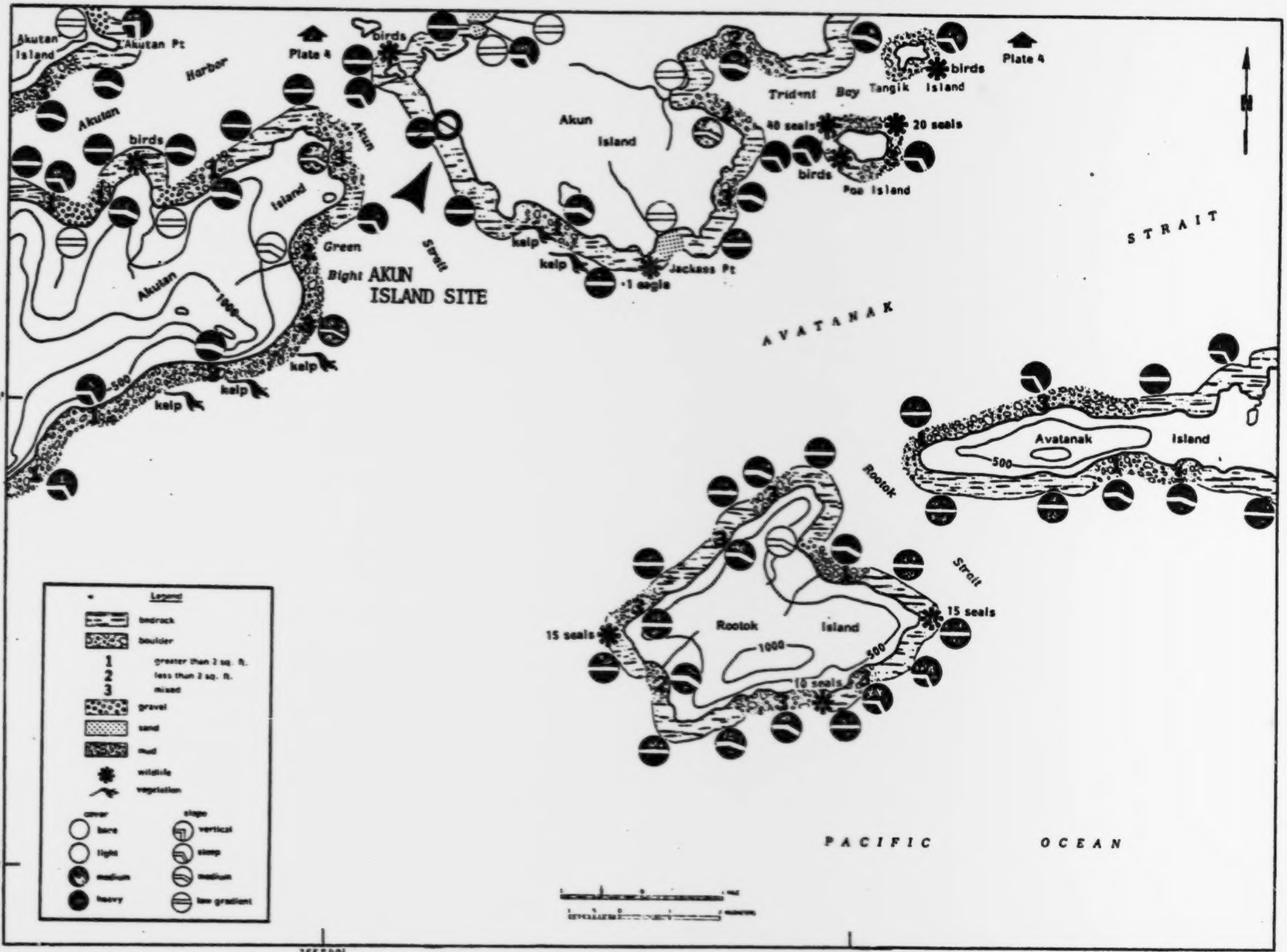
Akutan Island
Akutan Pt
Plate 4
Akutan Harbor
birds
Akutan Island
Akun Strait
Green Bight
AKUN ISLAND SITE
Akutan
1000
500
kelp
Akun Island
Trident Bay
Tangik Island
birds
Plate 4
40 seals
20 seals
birds
Poa Island
Jackass Pt
1 eagle
N
STRAIT
AVATANAK
Avatanak Island
500
Rootok Strait
Rootok Island
1000
500
15 seals
15 seals
10 seals
PACIFIC OCEAN
54° 05'
54° 00'
165° 40'
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
mi
kilometers

Fig. 25. Location of site at Akun Island.

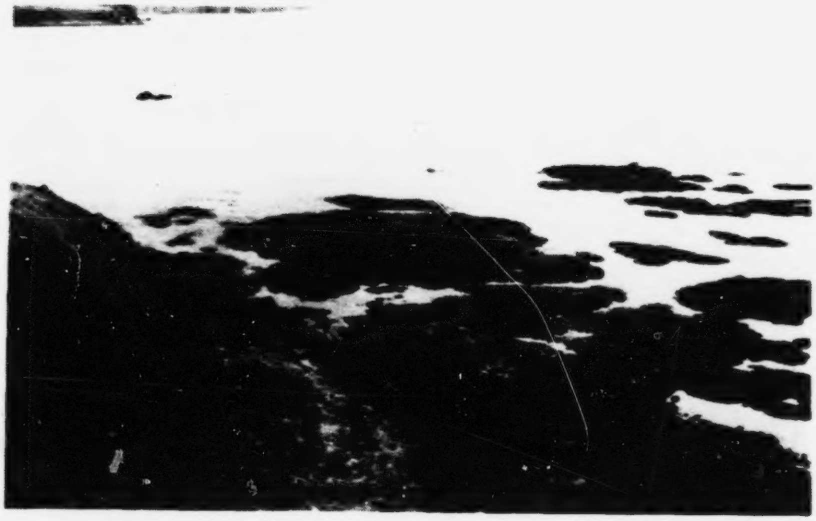

Fig. 26. Akun Island, Aleutian Islands. View of sampling area at low tide. 18 July 1975.

star Leptasterias hexactis was present in 11 quadrats. Algal cover was moderately heavy with no particular species or group dominant. We collected eight species of green algae, 13 species of red algae, and five species of brown algae. Many of the red algae species were finely branched, providing excellent habitat for the gastropods and nestling bivalves which were so abundant at this site.

1.14 Eider Point, Unalaska Island

The Eider Point site, located on the western shore of Unalaska Bay, Unalaska Island (Figs. 1 and 27; Table 1), consists of a low-gradient beach facing east. The substrate was composed of cobble, medium rocks, and boulders (Fig. 28). A steeper-gradient bedrock beach occurred north of the site below some high bluffs and a beach spit extended in a southerly direction from the site.

Four transects were established, one running at 50° magnetic and normal to the shoreline and three perpendicular to the normal transect. Sampling on the normal transect was systematic with a 1/16 m^2 quadrat placed every 5 m, beginning with 0 m at 2.17 m elevation and ending 35 m away at the zero m (zero tide) elevation. Horizontal transects were established by a stratifying procedure at the 0.61 m elevation contour (range 0.40 to 1.19 m) for Transect 2, the 0.06 m contour (range -0.12 to 0.52 m) for Transect 3, and the 0.06 m contour (range -0.15 to 0.24 m) for Transect 4. Transect 2 was selectively placed in an area in which Littorina sitkana was prevalent. Transect 3 fell in a zone dominated by Fucus. Transect 4 was placed in a zone dominated by Balanus cariosus and Alaria sp. (Fig. 29).

Sampling in the strata followed a randomized nested sampling design. Each stratum was subdivided into 20 contiguous 1 m^2 plots (m^2 plots will be called plots in the rest of this section) along each horizontal transect, and

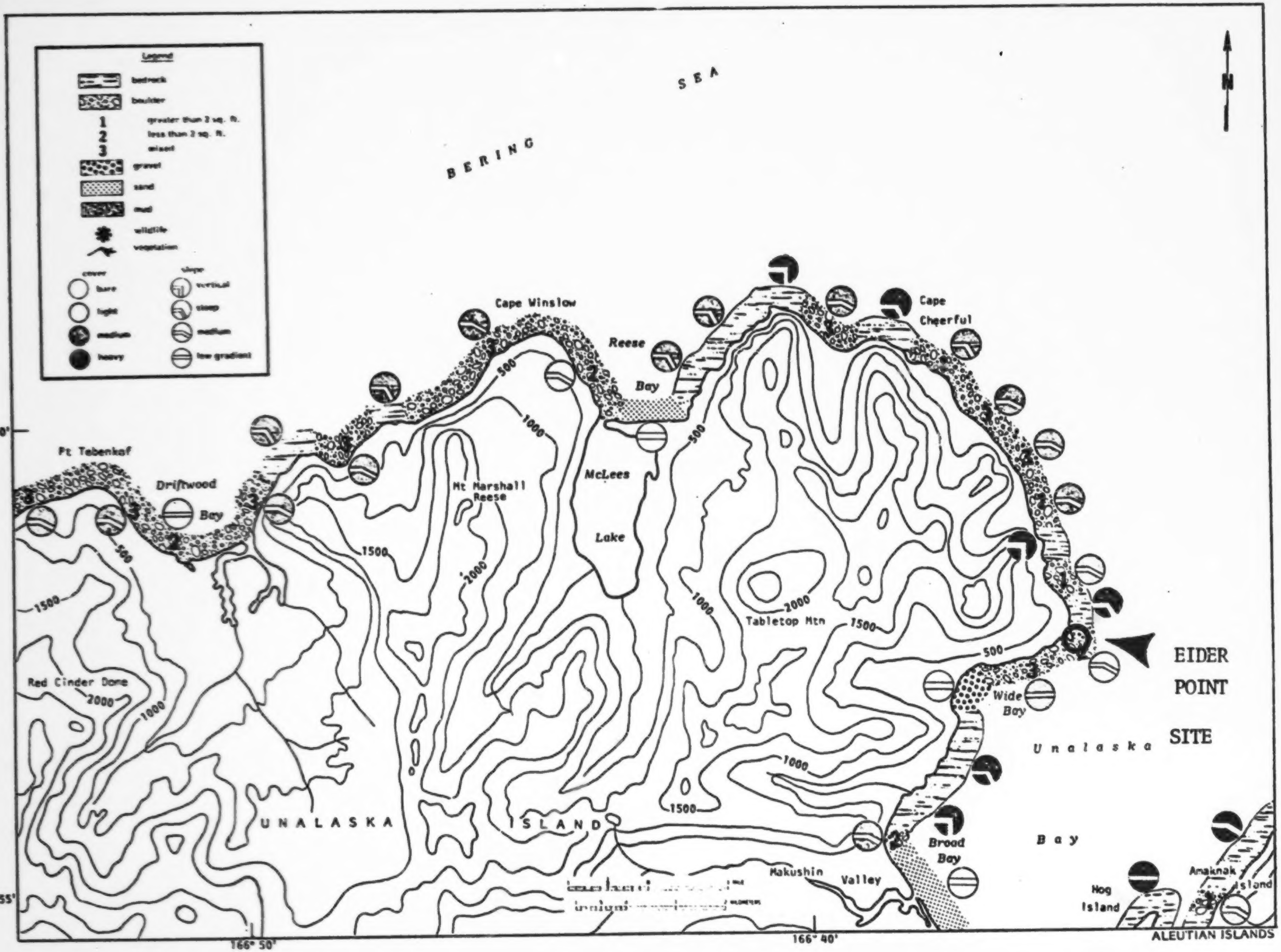
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
N
BERING SEA
Cape Winslow
Reese Bay
Cape Cheerful
54° 00'
Pt Tebenkof
Driftwood Bay
Mt Marshall Reese
McLees Lake
Tabletop Mtn
EIDER POINT SITE
Wide Bay
Unalaska Bay
Red Cinder Dome
UNALASKA ISLAND
Makushin Valley
Broad Bay
Hog Island
Amaknak Island
53° 55'
166° 50'
166° 40'
ALEUTIAN ISLANDS
500
1000
1500
2000

Fig. 27. Location of site at Eider Point.

Fig. 28. Site at Eider Point, Unalaska Island. 14 June 1976.

Fig. 29. View of the Alaria stratum and Transect 4 at Eider Point, Unalaska Island. 14 June 1976.

4 plots in each stratum were randomly chosen for sampling. Each plot was then subdivided into 16 quadrats 1/16 m^2 in size of which two quadrats were sampled randomly (except in transect 4 where one random quadrat was analyzed in one plot, three random quadrats in a second plot, and two random quadrats in each of the remaining plots were analyzed).

The biota was subdivided into four biomass categories for each stratum depending upon relative wet-weight biomass of the species or group. The dominant category I included each species or group that was represented in one or more samples and had a biomass exceeding 15 g wet weight. Category II included organisms which occurred in one or more samples and which were greater than 1.7 g wet weight and less than 15 g wet weight. Category III contained species and groups from 0.18 to 1.7 g wet weight. A small-sized category, defined as category IV, contained species and groups of 0.18 g wet weight or less. The greatest wet weight of any species or group established the biomass category for all other samples belonging to the same species or group.

Figure 30 shows the biomass by quadrat, elevation, and species. Total biomass is divided into four categories. The first quadrat along Transect 1 was the 5 m quadrat at 1.2 m elevation which contained only an egg case; the 10 m quadrat at 0.52 m elevation contained *Balanus glandula* and *Littorina sitkana*. The 15 m quadrat at the -0.01 m level contained *Strongylocentrotus droebachiensis* in category I. *Collisella pelta* contributed the highest biomass of the three species in category II. Four species were present in category III of which *Margarites beringensis* contributed the highest biomass. Four species were present in category IV of which *Eulalia quadrioculata* contributed the highest biomass.

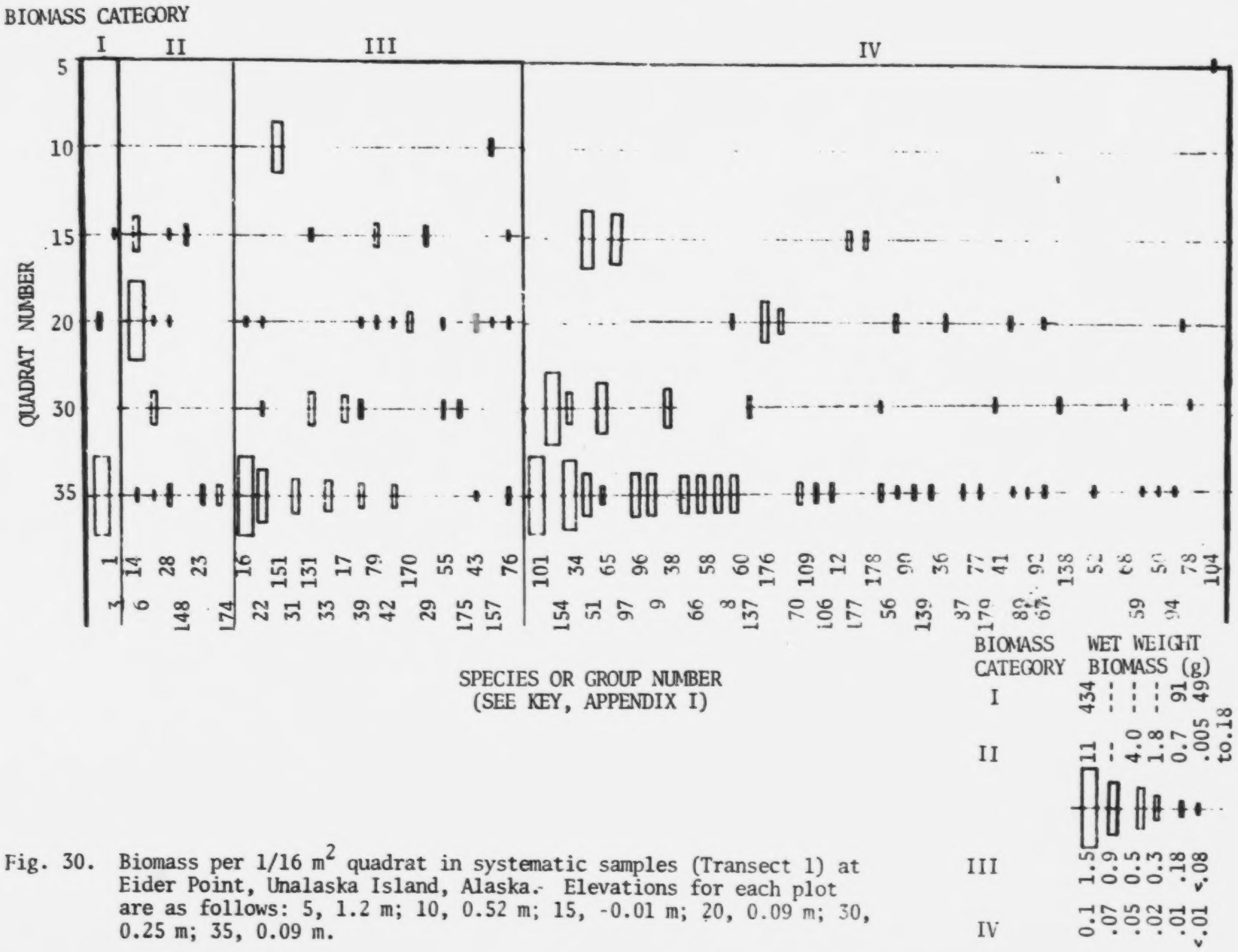

Fig. 30. Biomass per 1/16 m^2 quadrat in systematic samples (Transect 1) at Eider Point, Unalaska Island, Alaska. Elevations for each plot are as follows: 5, 1.2 m; 10, 0.52 m; 15, -0.01 m; 20, 0.09 m; 30, 0.25 m; 35, 0.09 m.

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The 20 m quadrat at 0.09 m elevation contained Balanus cariosus of category I, three species in category II of which Collisella pelta contributed the highest biomass for all quadrats of the transect for that category, ten species in category III of which Fabricia sabella contributed the highest biomass for the quadrat, and eight species in category IV of which Spongomorpha sp. contributed the highest biomass of the quadrat.

The 30 m quadrat at the 0.25 m level contained only Hiatella arctica for categories I and II. In category III there were six species of which Chlorophyta contributed the highest biomass. In category IV, Achelia chelata contributed the highest biomass of ten species.

The 35 m quadrat at the 0.09 m elevation contained the greatest number of species in each of the six quadrats. The highest biomass in any quadrat was represented by Balanus cariosus in the 35 m quadrat. Five species composed category II of which Nereis pelagica contributed the highest biomass. Eight species were represented in category III, with the highest biomass of the category for any quadrat represented by unidentified Zoantharia. Twenty-six species were present in category IV of which Monostroma sp. contributed the highest biomass of any species in this category.

The plateau area landward of 35 m was sampled by the 15 m, 20 m, and 30 m quadrats. No species was common to all three quadrats and only eight quadrat pairs shared a species. In contrast, the species-rich 35 m quadrat shared 16 of the total 42 species that occurred once or more within three quadrats landward of and nearest to 35 m and within the same elevation range.

Category I, which constituted 91% of the biomass in the Littorina-Balanus stratum of the nested random sample design, was represented by Balanus glandula and B. cariosus. Category II constituted 8% of the biomass of this stratum and was composed in descending order of biomass by Notoacmea scutum, Littorina sitkana, Collisella pelta, Nucella lima, Porifera, and Chthamalus dalli. Category III, which constituted 1% of the stratal biomass, was composed in descending order of biomass of Buccinum baeri, Endocladia sp., Fucus sp., Notoacmea persona, Pagurus beringanus, Cucumaria pseudocurata, Gigartina sp., and Pagurus sp. Category IV constituted <0.1% of the biomass of the stratum and was composed in descending biomass of Margarites beringensis, Typosyllis a. adamantea, Potamilla neglecta, Paramoera columbiana, Fabricia crenicollis, Terebellidae, and by other species ranging in wet weight from 0.002 to 0.02 g or those whose presence but not weight was recorded.

The 32 species and groups collected within the Littorina-Balanus stratum had a total wet weight of 477 g. Littorina sitkana was common to every quadrat within the stratum. Balanus sp. which constituted the highest biomass was common to seven of the eight quadrats collected in this stratum.

Category I constituted 91% of the wet-weight biomass of the Fucus stratum and was composed in descending biomass of Strongylocentrotus droebachiensis, Balanus cariosus, unidentified balanids, and unidentified coelenterates. Category II species constituted 8% of the wet weight biomass for this stratum and included Notoacmea scutum, Collisella pelta, Katharina tunicata, and Heliasteridae. Leptasterias sp., Balanus glandula, Chthamalus dalli, and Rhynchocoela constituted category III and 0.4% of the stratal biomass. The 28 category IV species and groups constituted 0.2% of the wet-weight biomass of the Fucus stratum. The five species and groups contributing most biomass (70%) to category IV were Echinoidea, Nereis sp., Porifera, Rhodophyta, and Margarites beringensis. The remaining 23 species and groups of category IV contributed 30% of the biomass to this category. A total of forty species or groups was identified for the *Fucus* stratum; they had a collective wet weight of 416 g.

The selection of the *Fucus* stratum at the 0.06 m (range -0.12 to 0.52 m) elevation level was based upon the presence of an obvious *Fucus* sp. band parallelling the shoreline at the site. However, of the eight quadrat samples taken in the *Fucus* stratum, only one quadrat contained *Fucus* sp. (0.15 g).

The distribution of mean wet-weight biomass in random quadrats within random plots by four biomass categories is shown for the *Alaria* stratum at Eider Point (Fig. 31). Category I constituted 98% of the wet-weight biomass of the *Alaria* stratum and is represented in descending biomass by *Balanus cariosus*, *Strongylocentrotus droebachiensis*, *Alaria crispa*, *A. c. taeniata*, and *Katharina tunicata*. There were 11 species and/or groups in category II with *Hiatella arctica* contributing the greatest biomass, followed by *Palmaria palmata* and *Mytilus edulis*. Category II contained 1.7% of the biomass in this zone. Category III comprised 34 species or groups and constituted 0.6% of the stratal biomass. *Nucella canaliculata* contributed greatest biomass, followed by tunicate fragments, *Pterosiphonia bipinnata*, *Leptasterias leptoderma*, *Monostroma fuscum*, *Mopalia ciliata*, and *Collisella* sp. Category IV consisted of 100 species or groups composing 0.1% of the total biomass for the stratum. The 10 organisms with greatest biomass were oligochaetes, *Nucella lima*, *Eulalia quadrioculata*, *Parallorchestes ochotensis*, *Phytichthys chirus*, polychaete fragments, *Asabellides sibirica*, Delesseriaceae fragments, *Eteone longa*, and *Typosyllis* sp.

Species common to each of the four plots in the *Alaria* stratum were *Balanus cariosus*, *Strongylocentrotus droebachiensis*, *Cirratulus cirratus*, Rhynchocoela, *Eulalia quadrioculata*, and Diptera larvae. Species present in

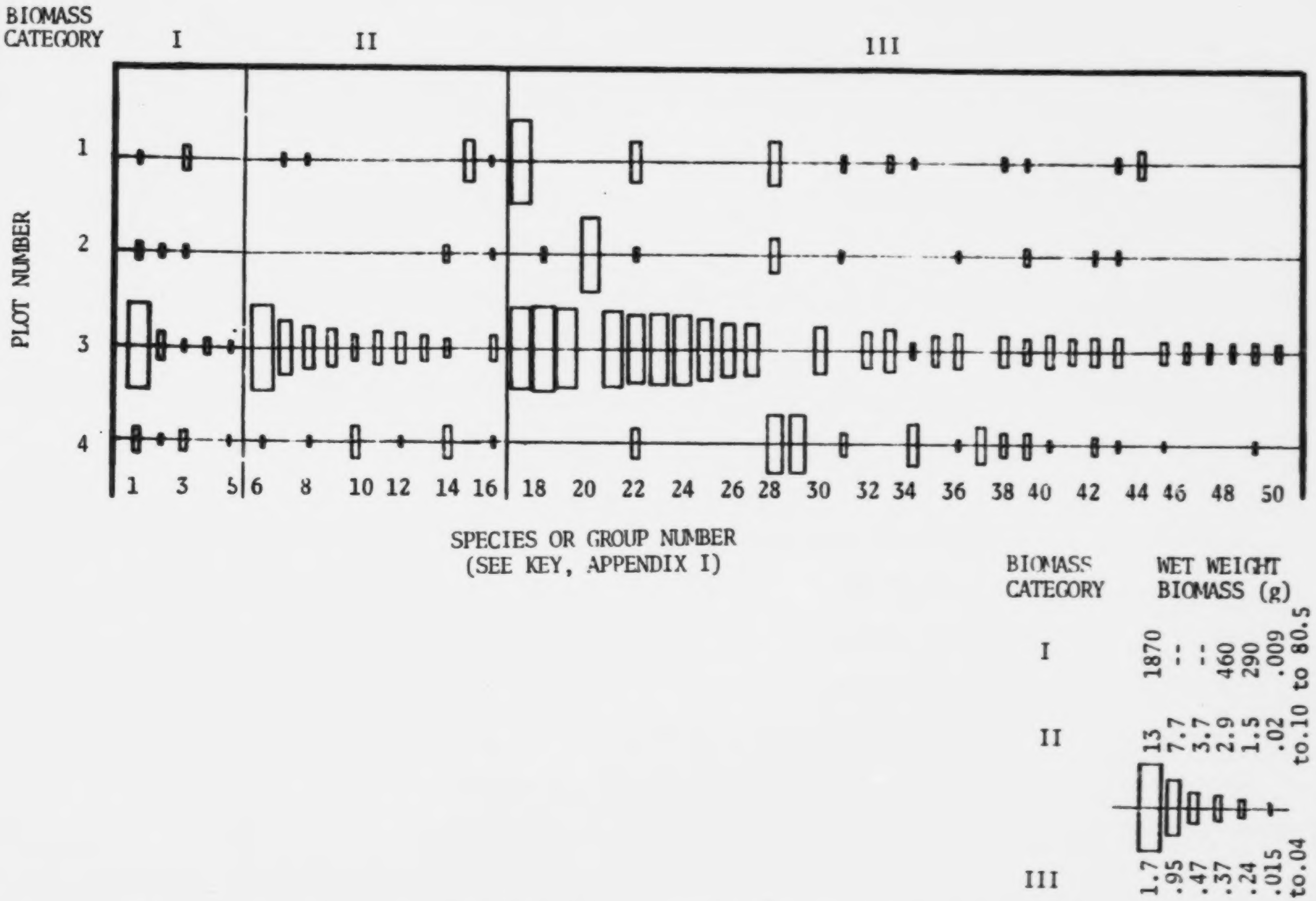

Fig. 31. Mean biomass per 1/16 m^2 quadrat in random m^2-plots along Transect 4 (the Alaria stratum) at Eider Point, Unalaska Island. Elevations for each plot are as follows: 1, 0.06 m; 2, 0.06 m; 3, 0.22 to 0.24 m; 4, 0.0 to -0.15 m. Only one quadrat sample was taken in plot #1; n = 2 for each of the other plots.

only three of the four plots for category I were Alaria crispa; for category II, Mytilus edulis and Collisella pelta; for category III, Nereis pelagica, Molgula oregonia, Cucumaria pseudocurata, Porifera, and Capitella capitata; and for category IV, Asabellides sibirica, Chone sp., nematodes, Margarites beringensis, Typosyllis alternata, and Polyplacophora. The 150 species or groups identified within the Alaria stratum had a collective wet weight of 10.2 kg.

Variability in species represented within quadrats in particular strata was large and required intensive sampling to adequately determine species composition. For example, in the Littorina-Balanus stratum of the random sampling design (range 0.4 to 1.19 m) 32 species or groups occurred compared to only two species in the 10 meter quadrat of Transect 1 at 0.52 m elevation of the systematic sample design. Inadequate sampling effort was also evident for the systematic sample at the Alaria stratum elevation (range of elevation within the Alaria stratum, -0.15 to 0.24 m). In the Alaria stratum, the 35 m quadrat of Transect 1 (at 0.9 m elevation) yielded 41 species or groups while eight quadrats placed along a transect perpendicular to Transect 1 produced 150 species or groups.

The Alaria stratum contained far more species and groups than were found elsewhere at the Eider Point site. Although this strata was the most seaward of the areas studied, the elevation of quadrats in the Alaria zone often equaled or exceeded that of plots located in more landward strata. For example, the elevation at the 15 m station was -0.01 m, whereas the elevation at the 20 m and 30 m sampling plots was 0.09 m. Quadrats in the Alaria stratum were between the elevation contours of -0.15 and 0.24 m and were often comparable in elevation to the more landward quadrats of the Fucus stratum between -0.12 and 0.52 m elevation.

The Eider Point site could be divided into two subcommunities or zones, one that depended more upon the seaward-landward axis than upon vertical elevations and a more landward zone that was dependent upon elevation. The first subdivision includes the more landward portion of the community on the "bench" of relatively similar elevations, as well as the more seaward quadrats represented by the *Fucus* stratum and the 15 m, 20 m, and 30 m quadrats of Transect 1 (Fig. 30). The second subdivision contains the quadrats of the *Alaria* stratum and the 35 m quadrat of Transect 1.

The dependency of the community upon the seaward-landward axis rather than elevation in the "bench" zone and more seaward locations may explain the occurrence of nearly the same numbers of species and groups in the 15 m, 20 m, and 30 m quadrats of Transect 1 as in the eight quadrats of the *Fucus* stratum lying perpendicular to the systematic sample transect.

1.15 Portage Bay, Makushin Bay

Portage Bay is about 1 mile wide and 4 miles long and forms a narrow arm of Makushin Bay on the northwest side of Unalaska Island (Figs. 1 and 32; Table 1). The site we sampled was a small, rocky point terminating in a cluster of large rocks (Figs. 33 and 34). Most of the bedrock in this area forms cliffs, thus has little biological cover. When lower-gradient rocky outcrops do occur, the biota covering them is extremely rich and varied. We collected 26 species of algae and 123 invertebrate species in our quadrat samples.

The lower intertidal zone with its well-developed growth of *Balanus cariosus* was of particular interest. *Balanus cariosus* cover was extremely heavy particularly below MLW (averaging 1424 individuals/m^2 and 14.9 kg/m^2) and their large, heavy plates provided evidence of an old population. Bare spots with isolated *B. cariosus* were observed (Fig. 35) but for the most part

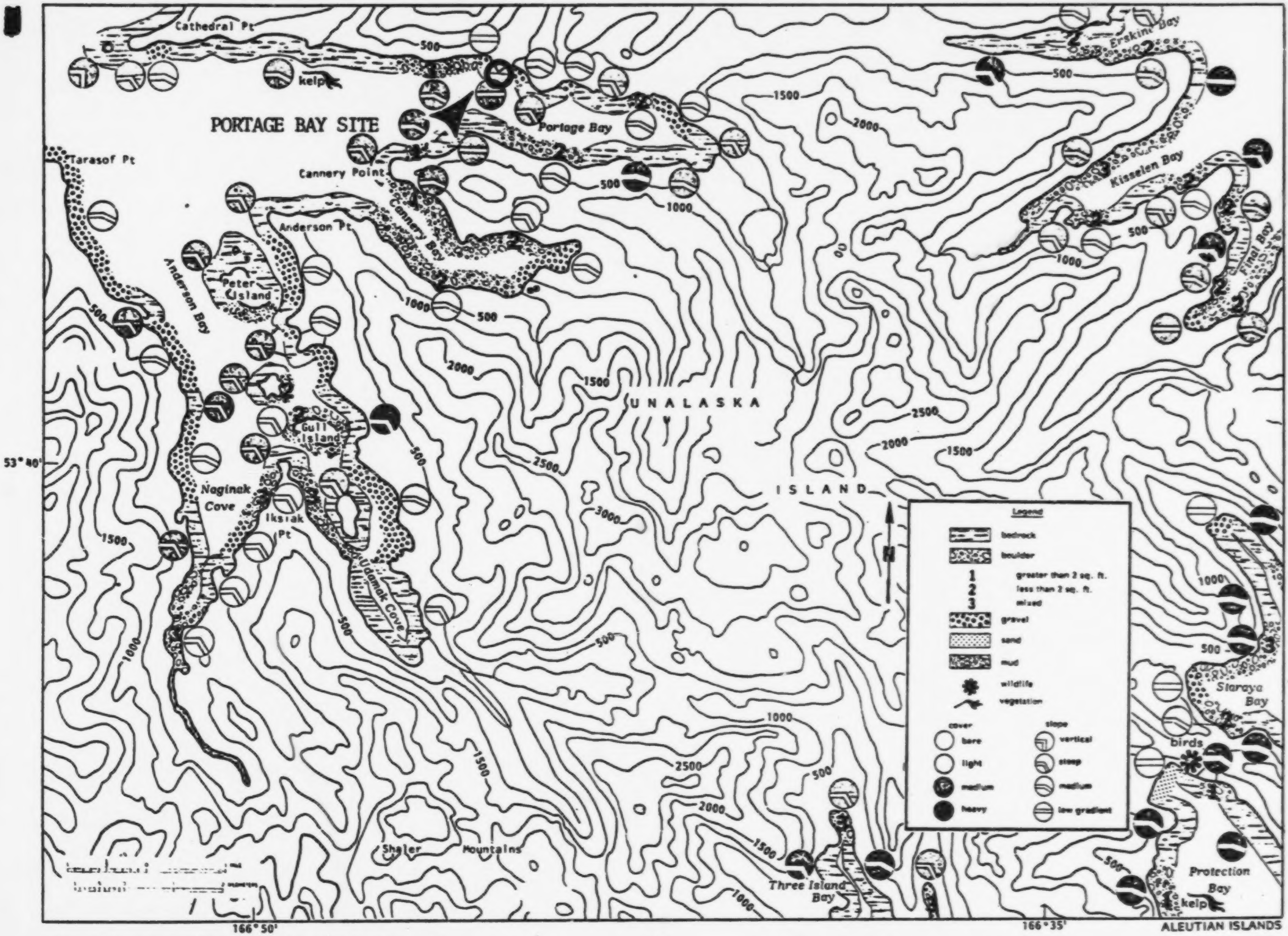
PORTAGE BAY SITE
Cathedral Pt
kelp
Portage Bay
Erskine Bay
Kisselen Bay
Final Bay
Tarasof Pt
Cannery Point
Cannery Bay
Anderson Pt
Anderson Bay
Peter Island
Gull Island
Naginak Cove
Iksiak Pt
Udamak Cove
UNALASKA
ISLAND
Staraya Bay
birds
Protection Bay
kelp
Three Island Bay
Shaler Mountains
N
53°40'
166°50'
166°35'
ALEUTIAN ISLANDS
500
1000
1500
2000
2500
3000
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 32. Location of site at Portage Bay.

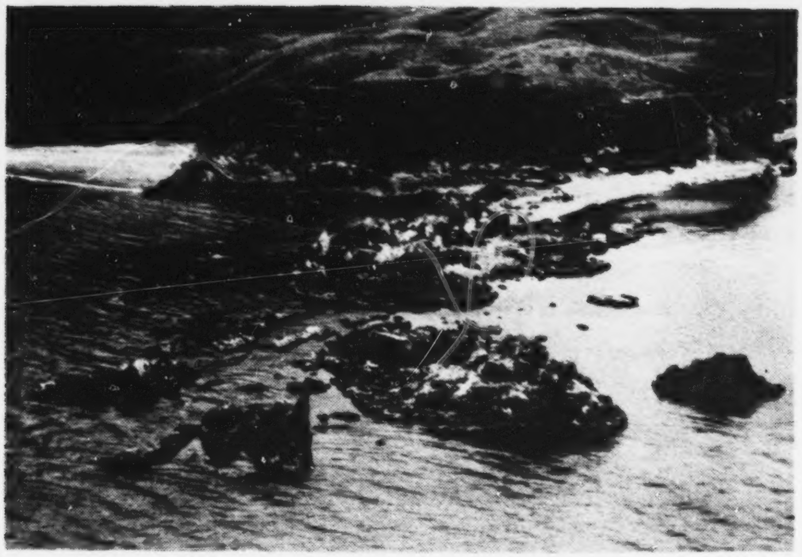

Fig. 33. Aerial view of the sampling site at Portage Bay, Unalaska Island, Aleutian Islands. Rocky outcrops of this type are uncommon in Portage Bay. 25 July 1975.

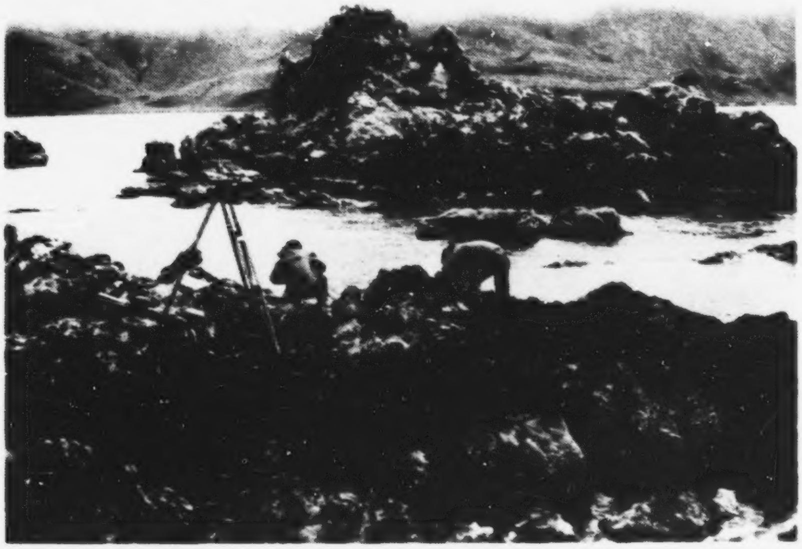

Fig. 34. Portage Bay sampling area. 13 August 1975.

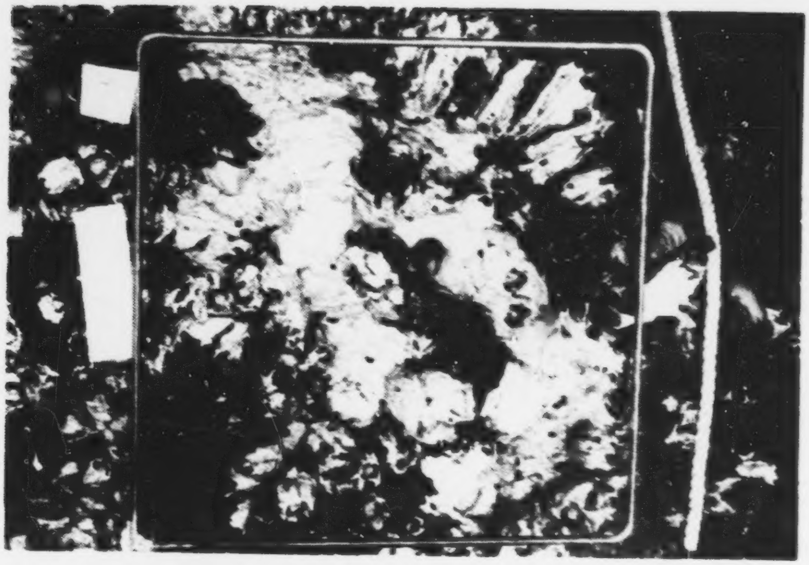

Fig. 35. Typical invertebrate assemblage in Balanus cariosus zone at Portage Bay site; note old, crowded barnacles. 25 July 1975.

Fig. 36. View of sampling area showing part of transect line, zonation study site and plethora of workers. Inadvertent trampling by the sampling crew on this small, heavily-covered area had a devastating effect on barnacles and mussels. 25 July 1975.

their density was remarkably uniform. Trampling by many people concentrated in a small area proved to be a major disturbance to the barnacle population (Fig. 36). This was evident when we returned to the area in August when we saw not only cleared, uncolonized patches of bare rock where our quantitative samples had been removed but also remains of crushed B. cariosus that had been trampled by the overabundance of workers. In August, we concentrated our efforts higher on the beach in order to complete our survey of the site and to avoid the disturbed low zone.

Three species of barnacles, Balanus glandula, B. cariosus, and Chthamalus dalli, were found in our quadrat collections. Although B. glandula was numerically dominant in more quadrats than were the other two species (Table 10), B. cariosus dominance in the low zone was strikingly apparent because of its greater biomass.

Predation on barnacles appeared to be minimal. Potential predators such as Nucella spp. and sea stars were not abundant. Leptasterias hexactis, a small sea star, was present, but it was probably not an important source of barnacle mortality. Mature Balanus cariosus probably experience only minor predation pressure due to their large size. One possible major source of predation to large Balanus was the black oystercatcher, Haematopus bachmani, which was abundant and which we observed eating barnacles.

The extensive barnacle cover provided a shelter for two diverse groups. We collected 12 families and 26 species of polychaetes, many of which were found in the interstices between barnacles. We also collected an unusually large number of gastropods (17 species) of which many individuals measured only a few millimeters in length. These gastropods also take shelter in the interstices between barnacles or in empty barnacle tests.

Table 10. Constancy, dominance, and tidal range of three species of barnacles collected at Portage Bay, Alaska. Constancy is the proportion of quadrats that contained the barnacle. Dominance is the proportion of quadrats in which the barnacle was numerically dominant over other barnacles. There were two quadrats (at 0.67 m and 1.62 m) where no barnacles were found. (a) and (b) refer to the ranges of elevations of samples used in calculating constancy and dominance, respectively.

Species	Constancy	Dominance	Tidal range (m)
Balanus glandula	29/42	24/42	(a) 0.46 to 1.25 (b) 0.73 to 1.25
Balanus cariosus	22/42	13/42	(a) 0.09 to 0.91 (b) 0.09 to 0.79
Chthamalus dalli	5/42	5/42	(a) 0.40 to 0.82 (b) 0.40 to 0.82

The littorine snail *Littorina sitkana* was found in almost every quadrat, and was most abundant in the upper intertidal zone. Although the abundance of *L. sitkana* along our transect was adequately assessed in our quadrat collections, even greater numbers were observed in damp shaded areas several meters shoreward of our sampling area (Fig. 37).

1.16 Crooked Island

Crooked Island is a member of the Walrus Island group in northeastern Bristol Bay (Figs. 1 and 38; Table 1). Of all the sites we sampled quantitatively, Crooked Island was the one most influenced by freshwater runoff from the rivers of the Alaska Peninsula. In this area, a large shoal extends well offshore. The Walrus Islands lie about 16 km offshore from the entrance of Togiak Bay. The head of Togiak Bay is emersed southward to a distance of 8 to 10 km at low tide (U.S. Coast Pilot #9, 1964). The influence of the silt-laden freshwater from the Togiak River extended to Crooked Island. A thin layer of very fine silt covered everything, and the low-lying areas between bedrock outcrops were filled with mud. Crooked Island is also subject to the effects of sea ice. The mean southern limit of sea ice is shown at or south of Crooked Island from 1 November through 31 May on the charts of Wise and Searby (1977).

We sampled a beach composed of gravel and small angular rock and a flat bedrock bench seaward of the beach (Fig. 39). A single transect was placed perpendicular to the shore and sampled every 5 m (Table 1); all but four quadrats fell below mean tide level. We made qualitative observations of the biota in the muddy areas.

Fucus distichus, in poor condition with ragged fronds and generally desiccated appearance, had the greatest biomass in half of the quadrats; one quadrat contained no organisms; and *Zostera marina* occurred in one quadrat

Fig. 37. Concentration of littorine snails along supralittoral fringe at Portage Bay sampling site. 25 July 1975.

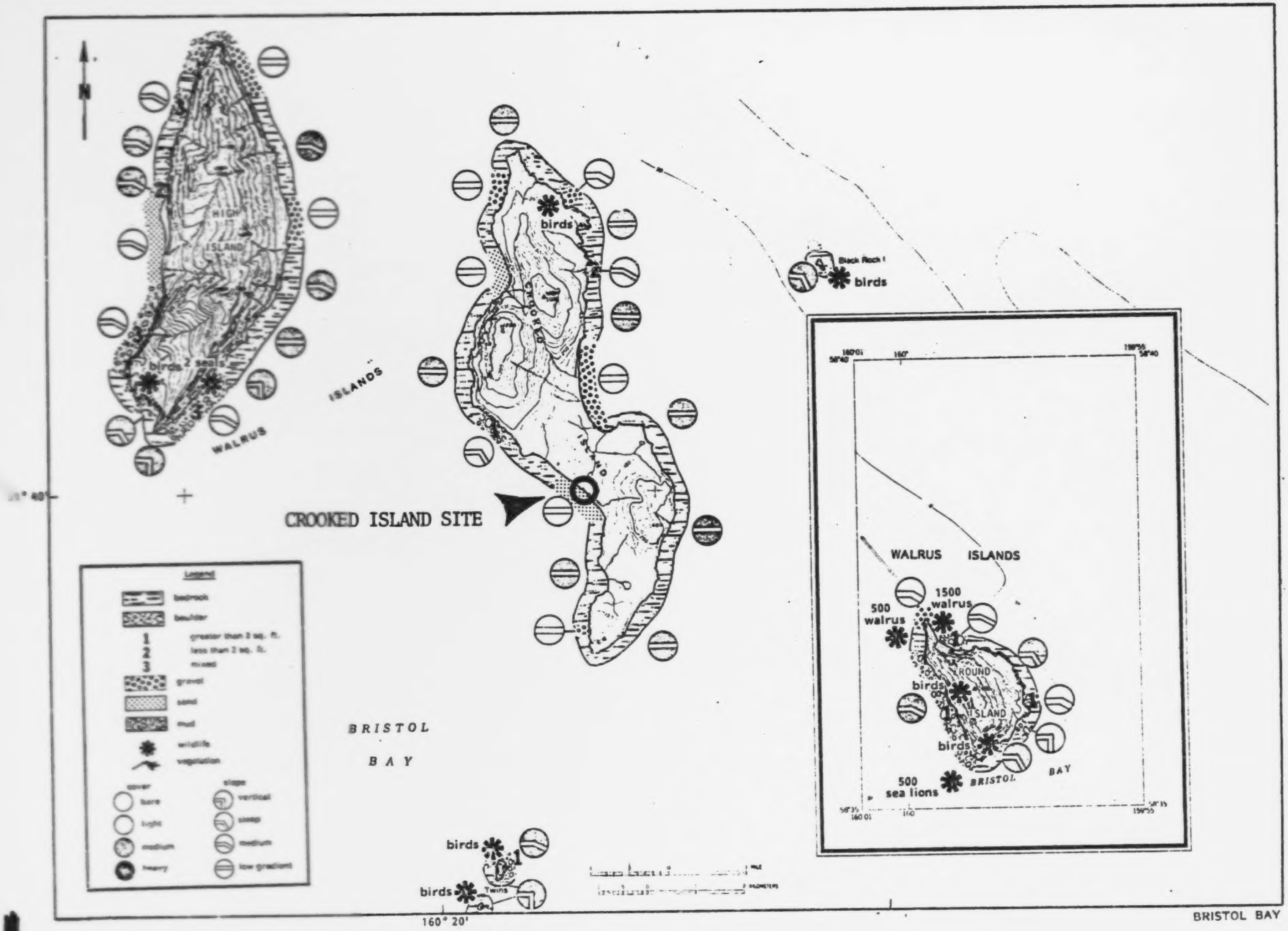
N
HIGH ISLAND
birds
2 seals
WALRUS ISLANDS
CROOKED
birds
CROOKED ISLAND SITE
Black Rock I
birds
BRISTOL BAY
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
birds
birds
Twins
MILE
KILOMETERS
160° 20'
WALRUS ISLANDS
500 walrus
1500 walrus
ROUND ISLAND
birds
birds
500 sea lions
BRISTOL BAY
160°01'
160°
159°55'
58°40'
58°35'
BRISTOL BAY

Fig. 38. Location of site at Crooked Island.

205

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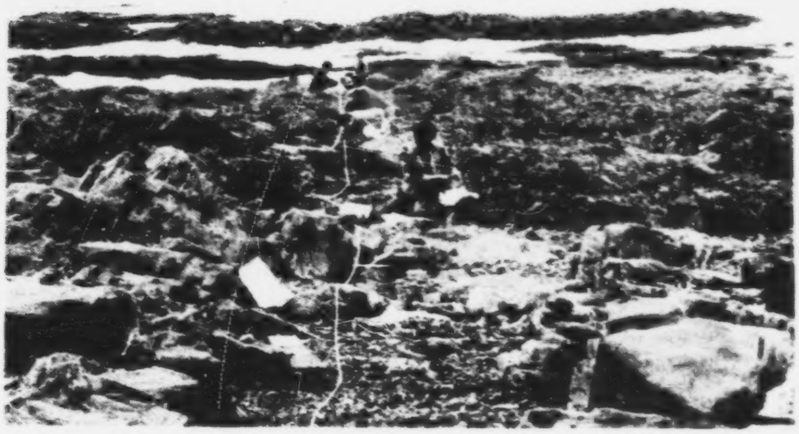


Fig. 39. Looking seaward along the transect line at Crooked Island. 20 July 1975.

and was dominant there. Dominance in the other quadrats was distributed fairly evenly among seven other species (Table 11). Barnacles Chthamalus dalli and Balanus glandula were the most abundant and most contagiously distributed of the large invertebrates. Although we made no observations of feeding relationships, the barnacles were probably being preyed on by Nucella lima, which was relatively common in the plots (Table 11).

Mytilus edulis occurred more frequently than any other species along the transect but its density and biomass were low. In our collection, every Mytilus except one was less than 1.5 cm in length. Apparently Mytilus settles in the lower intertidal zone at this site but most do not survive to a large size. These small Mytilus are probably also preyed on by Nucella. The muddy areas contained many crabs, polychaetes, large burrowing anemones, and Telmessus cheiragonus.

1.17 Cape Peirce

Cape Peirce is a rock headland southeast of Cape Newenham on the northeastern shore of Bristol Bay (Figs. 1 and 40; Table 1). Most of the shore at Cape Newenham is bordered by steep cliffs on which large seabird colonies (especially those of black-legged kittiwakes) occurred. Our sites were on a low elongated bedrock mound surrounded by sand and gravel beach and on an adjacent vertical rock wall (Figs. 41 and 42). We sampled the mound by haphazardly tossing a 1/16 m^2 frame on it and removing the organisms within the frame. The vertical face was sampled with the arrow method. All quadrats but one fell below mean tide level.

The species assemblages at the two sites (mound and arrow) were similar. Of the species accounting for most of the biomass at Cape Peirce, only Balanus glandula was not present at both sites. Its absence from the

Table 11. Statistics for the 11 species of algae and invertebrates showing the greatest average rank on the transect at Crooked Island, Bristol Bay.

Species	Average rank[a]	Wet weight (g)[b]	Frequency of occurrence[c]	Abundance[d]	Dominance[e]
Fucus distichus	7.2	103.3, 1.5-482.1	0.61	--	9/18
Chthamalus dalli	5.9	6.7, 0.9-90.8	0.61	152, 37-1190	3/18
Mytilus edulis	5.0	0.4, 0.002-4.6	0.67	6, 1-36	0
Nucella lima	4.8	6.7, 0.08-30.9	0.5	2, 1-16	2/18
Scytosiphon lomentaria	4.1	0.8, 0.06-9.8	0.44	--	0
Balanus glandula	4.1	7.9, 0.03-91.4	0.5	128, 4-1383	2/18
Odonthalia floccosa	3.2	3.6, 0.4-36.4	0.33	--	1/18
Littorina sitkana	2.9	10.1, 0.6-49.2	0.44	54, 1-237	1/18
Corallina sp.	2.5	1.0, 0.08-7.7	0.17	--	0
Pagurus h. hirsutiusculus	2.3	0.3, 0.2-7	0.28	2, 1-9	1/18
Ahnfeltia plicata	1.9	1.2, 0.7-31.1	0.17	--	1/18

a. Rank of wet weight of each species averaged over 18 quadrats (1, lowest weight). Species listed in order of decreasing rank.

b. Median and range of wet weights per 1/16 m^2 in all quadrats where the species was found.

c. Proportion of quadrats that contained the species.

d. Animals only; median and range of number of individuals per 1/16 m^2 in all quadrats where the species was found.

e. Proportion of quadrats in which the species was among those making up 50% of the wet weight. Summation in each quadrat began with the heaviest species.

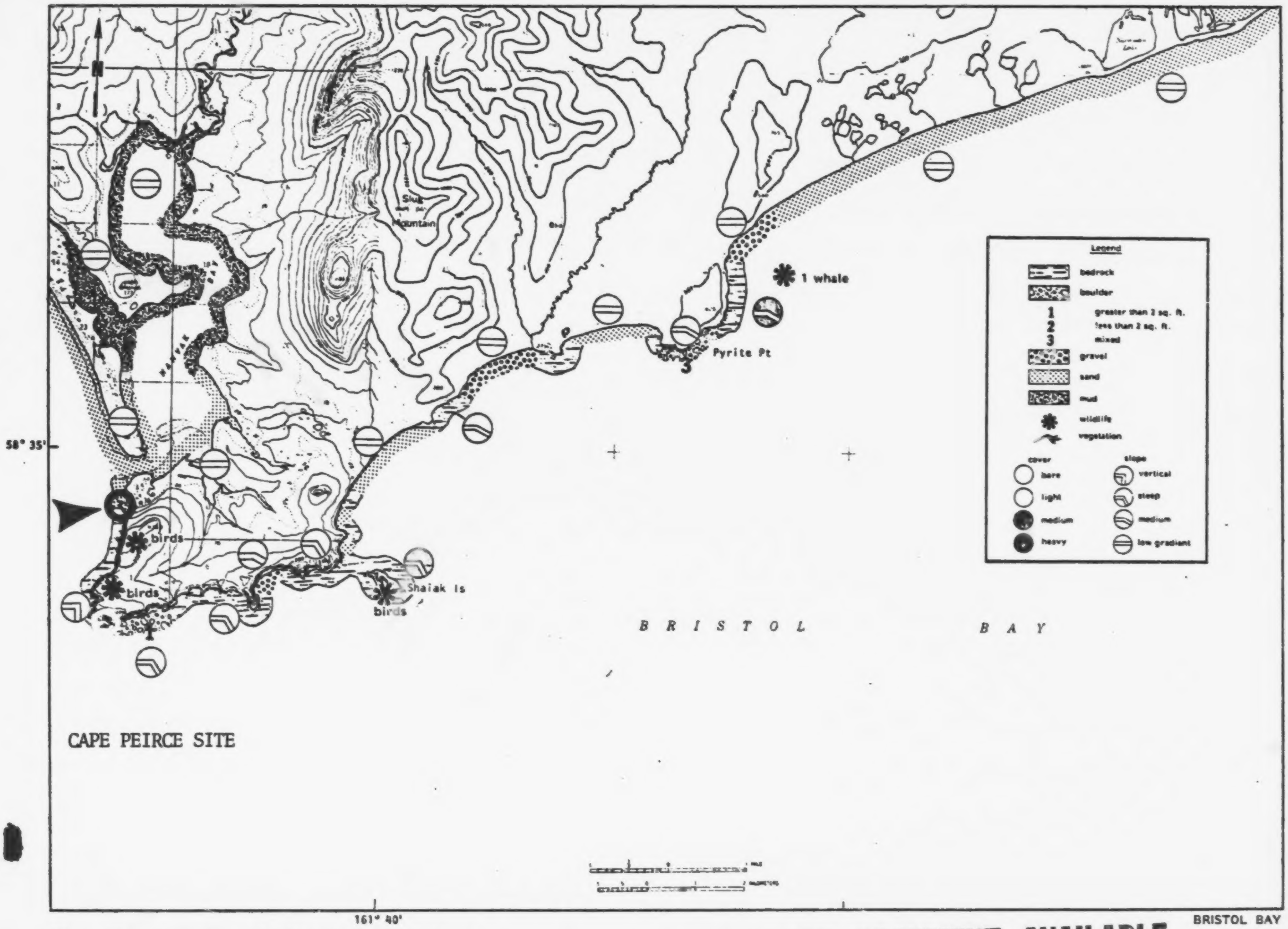

Sluk Mountain
1 whale
Pyrite Pt
birds
birds
Shaiak Is
birds
B R I S T O L B A Y
58° 35'
161° 40'
CAPE PEIRCE SITE
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 40. Location of site at Cape Peirce.

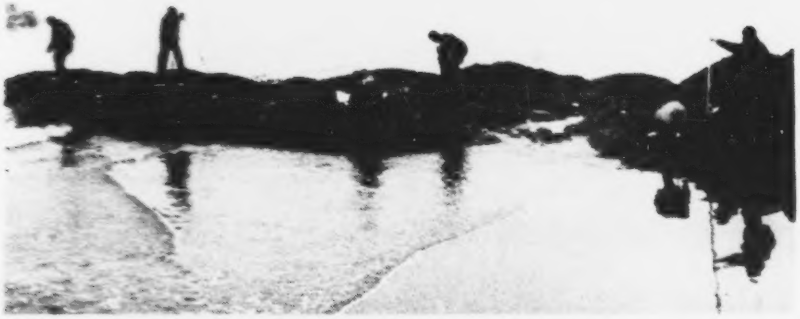


Fig. 41. View of the bedrock mound site at Cape Peirce; sampling here was non-systematic. 21 July 1975.

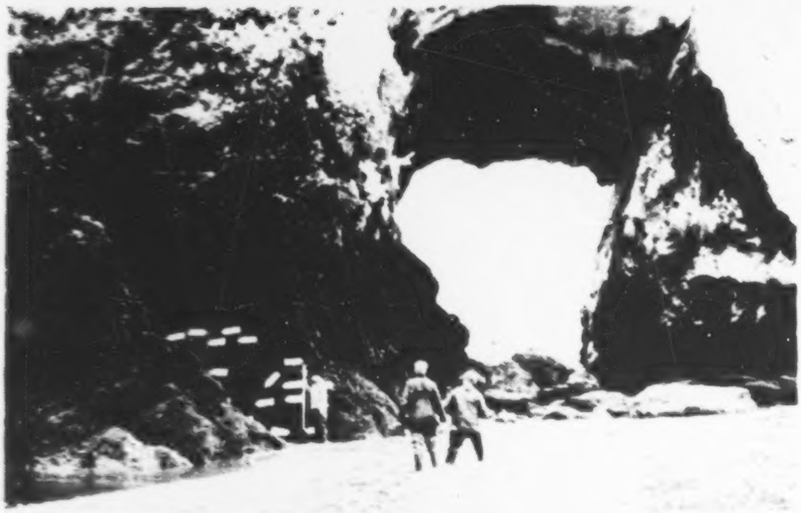


Fig. 42. View of zonation study (arrow) site at Cape Peirce. 21 July 1975.

arrow site was probably an artifact of sampling because sample size there was small (n = 4). Samples from both sites were lumped for the analysis.

At Cape Peirce, as at Crooked Island, Fucus distichus and Chthamalus dalli had the greatest average rank (based on wet weight) of all species present (Table 12). About half of the ten species with the greatest average rank were common to both localities. Corallina sp., Ahnfeltia plicata, Odonthalia floccosa, Nucella lima, and Pagurus h. hirsutiusculus were completely absent from our samples at Cape Peirce. Conversely, Melanosiphon intestinale, Halosaccion glandiforme, and Chironomidae were absent from the Crooked Island samples. It remains to be determined whether the difference in species composition between localities is real or is merely an artifact resulting from the relatively small sample size and difference in the size of the area sampled (the area sampled at Cape Peirce was smaller than that at Crooked Island). Because of the magnitude of the change (from absence at one locality to near dominance in biomass at the other), our data probably reflect real differences in the abundances of these species between localities.

Among the primary competitors for space at both localities, F. distichus showed a substantially lower biomass at Cape Peirce; B. glandula was slightly less abundant at Cape Peirce than at Crooked Island and C. dalli was slightly more abundant at Cape Peirce than at Crooked Island (Table 11). Balanus balanoides was recorded in two quadrats (abundances, 913 and 287 individuals per quadrat) at Cape Peirce and was not recorded at Crooked Island. Mytilus edulis was small and low in abundance in quadrats at both localities and occurred less frequently at Cape Peirce than at Crooked Island.

The chief carnivore at Crooked Island, Nucella lima, was absent from samples taken at Cape Peirce. The consequences of its absence or reduced abundance at Cape Peirce are impossible to determine with our data.

Table 12. Statistics for the 10 species of algae and invertebrates showing the greatest average rank in quadrats at Cape Peirce, Bristol Bay.

Species	Average rank[a]	Wet weight (g)[b]	Frequency of occurrence[c]	Abundance[d]	Dominance[e]
Fucus distichus	6.2	6.7, 0.2-183	0.76	-	5/17
Chthamalus dalli	5.6	21, 0.4-120.2	0.76	311, 9-1382	6/17
Melanosiphon intestinale	3.8	1.8, 0.04-10	0.53	-	1/17
Littorina sitkana	3.7	0.2, 0.01-0.08	0.94	27, 2-86	0
Balanus glandula	3.6	5.2, 0.01-84.8	0.47	30, 1-491	3/12
Halosaccion glandiforme	3.3	2.5, 0.04-66.3	0.65	-	1/17
Mytilus edulis	2.2	0.06, 0.001-0.2	0.59	8, 1-38	0
Scytosiphon lomentaria	1.1	0.08, 0.3-4.3	0.18	-	0
Enchytraeidae	0.9	0.005, 0.001-0.02	0.35	22, 3-53	0
Chironomidae (larvae)	0.9	0.001, 0.001-0.008	0.41	5, 1-13	0

a. Rank of wet weight of each species averaged over 17 quadrats (1, lowest weight). Species listed in order of decreasing rank.

b. Median and range of wet weights per 1/16 m^2 in all quadrats where the species was found.

c. Proportion of quadrats that contained the species.

d. Animals only; median and range of number of individuals per 1/16 m^2 in all quadrats where the species was found.

e. Proportion of quadrats in which the species was among those making up 50% of the wet weight. Summation in each quadrat began with the heaviest species.

Differences in the abundance of the preferred prey (barnacles and mussels) of *Nucella* between localities are not consistent with predictions based on difference in the abundance of *Nucella*.

2. The Pribilof Islands

The shores of the Pribilof Islands (lat. 57°N, long. 170°W) provide the only littoral habitat in the St. George Basin (the area in the southeastern Bering Sea extending northwest of Unimak Island to and including St. George Island (Figure VI-1-1 in Environmental Research Laboratories, 1977). Berg (1977, Fig. 1) shows a map of the proposed oil and gas lease areas in this basin.

We studied seven sites on three of the four islands which constitute the Pribilof group (Fig. 43): three sites on St. Paul, three on St. George, and one on Otter Island (Table 13); Walrus Island, the smallest, was not studied. Species in those samples collected at our study sites in the Pribilof Islands are listed in Appendix IIC. Appendix III is a list compiled by N. Calvin of algae collected by divers in the Pribilof Islands.

2.1 St. Paul Island

2.1.1 English Bay--The English Bay sampling site is a low-gradient boulder beach on the south shore of St. Paul Island midway between Zapadni and Tolstoi Points (Figs. 43 and 44). The study area is within sight of the village of St. Paul and is adjacent to one of the larger fur seal rookeries. At the time of our sampling on 10 June 1976, bull fur seals were just beginning to arrive from southern waters for the annual breeding cycle. This site, more than any other which we visited in the Bering Sea and Norton Sound, exhibited striking effects of ice scour on the intertidal biota. Much of the sampling transect traversed an area of flat-topped boulders up to 1 m in diameter, which had been scoured by ice. Rich assemblages of biota occurred in the protected interstices beneath and between boulders, but upper surfaces that had been abraded by ice were nearly bare. Thick crusts of coralline algae occurred in crevices.

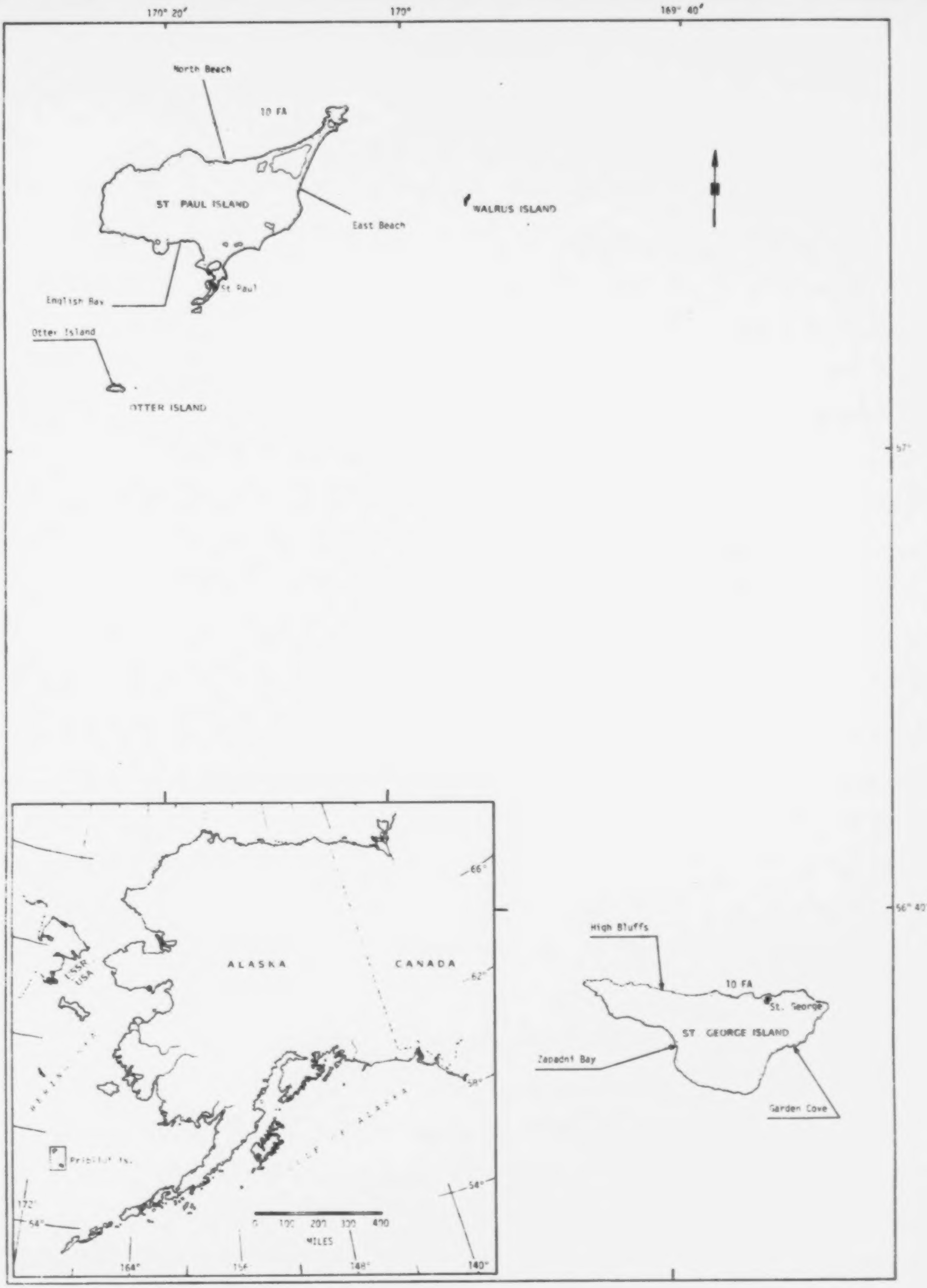

170° 20'
170°
169° 40'
North Beach
10 FA
ST PAUL ISLAND
East Beach
WALRUS ISLAND
English Bay
St Paul
Otter Island
OTTER ISLAND
57°
56° 40'
High Bluffs
10 FA
St. George
ST GEORGE ISLAND
Zapadni Bay
Garden Cove
ALASKA
CANADA
USSR
USA
BERING SEA
GULF OF ALASKA
Pribilof Is.
66°
62°
58°
54°
172°
54°
0 100 200 300 400
MILES
164°
156°
148°
140°

Fig. 43. Location of study sites in the Pribilof Islands.

Table 13. Pertinent sampling information for seven sites in the Pribilof Islands.

Site	Lat.(N)	Long.(W)	Substrate Type	Dates Sampled	Tidal Range Sampled (cm)	Sampling[a] Method	Quadrat Surface (cm)	Sample[b] Size
Pribilof Islands								
St. Paul Island								
English Bay	57°9.3'	170°18.8'	Boulder	10 Jun 76	-34 to +195	Transect (1)	625	14
North Beach	57°12.8'	170°12'	Sand	12 Jun 76	0 to +60	Selected (2)	100[c]	2
East Beach	57°11.4'	170°9.2'	Sand	12 Jun 76	0 to +60	Selected (2)	100[c]	2
Otter Island	57°02.9'	170°23.6'	Boulder-bedrock	16 Aug 75	+8 to +242	Transect(2), Arrow	625	34(T), 5(A)
				12 Jun 76	-34 to +284	Transect(2), Random	625	17(T), 7(R)
St. George Island								
Garden Cove	56°33.8'	169°31.1'	Bedrock	9 Jun 76	+3 to +25	Random, Transect(2), Selected	625	22(T), 8(S)
			Sand		+12.2 to +250	Selected	100[c]	4
High Bluffs	56°36.4'	169°49.9'	Boulders	13 Jun 76	-21 to +372	Transect(2) Selected	625	16(T), 6(S)
Zapadni Bay	56°34.0'	169°40.0'	Boulder-bedrock	15 Aug 75	-33 to +156	Transect(2)	625	44

a. Number in parentheses indicates number of transects, arrow stations, etc. if more than one was established.

b. Letter in parentheses indicates sampling method: T, systematic Transect; A, arrow; R, random; S, selected.

c. Samples 100 cm^2 in area were cores 10 cm deep.

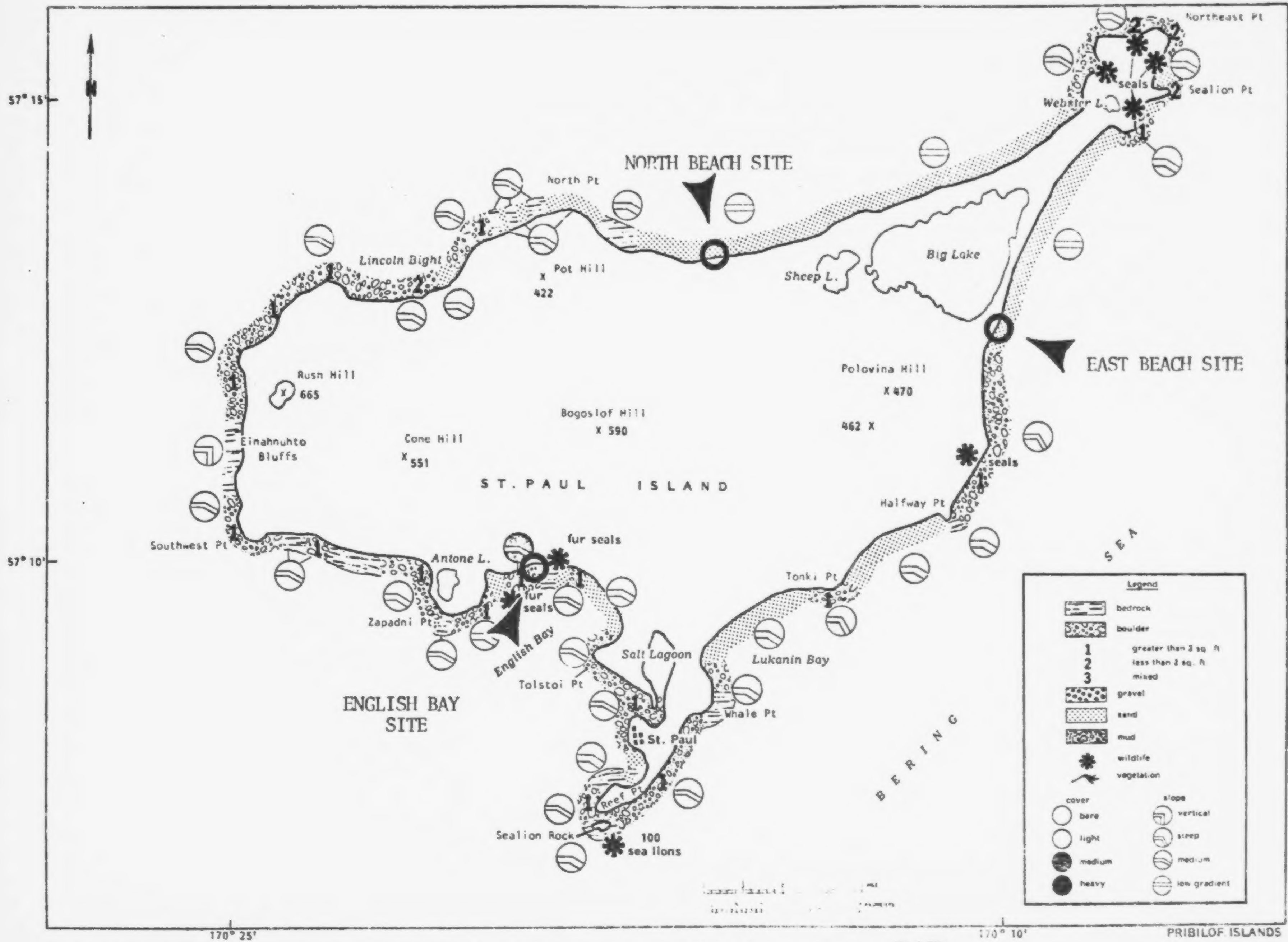

N
57° 15'
57° 10'
170° 25'
170° 10'
NORTH BEACH SITE
EAST BEACH SITE
ENGLISH BAY SITE
Northeast Pt
Sealion Pt
seals
Webster L.
North Pt
Lincoln Bight
Pot Hill 422
Sheep L.
Big Lake
Rush Hill 665
Polovina Hill X 470
462 X
Bogoslof Hill X 590
Cone Hill X 551
Einahnuhto Bluffs
ST. PAUL ISLAND
seals
Halfway Pt
Southwest Pt
fur seals
Antone L.
fur seals
Zapadni Pt
English Bay
Tolstoi Pt
Salt Lagoon
Tonki Pt
Lukanin Bay
Whale Pt
St. Paul
Reef Pt
Sealion Rock
100 sea lions
BERING SEA
Legend
bedrock
boulder
1 greater than 2 sq. ft
2 less than 2 sq. ft
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
PRIBILOF ISLANDS

Fig. 44. Location of the three St. Paul Island sites: English Bay, North Beach, and East Beach.

We collected six 1/16 m^2 samples spaced 5 m apart along a single transect. The gradient of the transect was slight, with a range of only +0.15 to -0.34 m. Turtonia occidentalis was the most abundant animal at upper and mid-tide levels. At tidal elevations below -0.1 m, luxuriant growths of Alaria sp. and Odonthalia/Rhodomela were the dominant algae, and between the -0.25 and -0.34 m tide levels Laminaria longipes grew in profusion from between boulders. The stipes had been sheared off at the level of the tops of the boulders, giving the appearance of a mowed lawn.

2.1.2 North Beach and East Beach--Two similar exposed sand beaches were sampled on each side of Northeast Point, St. Paul Island, on 12 June 1976 (Figs. 43 and 44). The North Beach site was about midway along a 9 km beach on the north side of St. Paul; the East Beach site was midway along a similar 5 km beach on the northeast side of the island. They were characterized by hard-packed black volcanic sand with no organisms evident except occasional amphipods in the surf zone. Two 1-liter cores were collected at each beach, one at the water's edge at low tide, and the other at the mid-tide level (approximately +60 cm). At the North Beach site, 7 mysids (Archoeomysis grebnitzkii) were found in the lower sample; the sample from higher on the beach contained two mysids and fragments of Porphyra sp. At the East Beach site there were no macroorganisms in the lower sample and only a few fragments of red algae in the upper sample.

Hundreds of kilometers of similar high-energy sand beaches occur along the Bering Sea coast; most are probably as depauperate of macrofauna as these at St. Paul Island because of unstable substrate, vulnerability to physical abrasion by ice, and exposure to storm surf. These physical conditions allow colonization by very few species. Exposed sand beaches probably contribute little to productivity of nearshore waters in the Pribilofs.

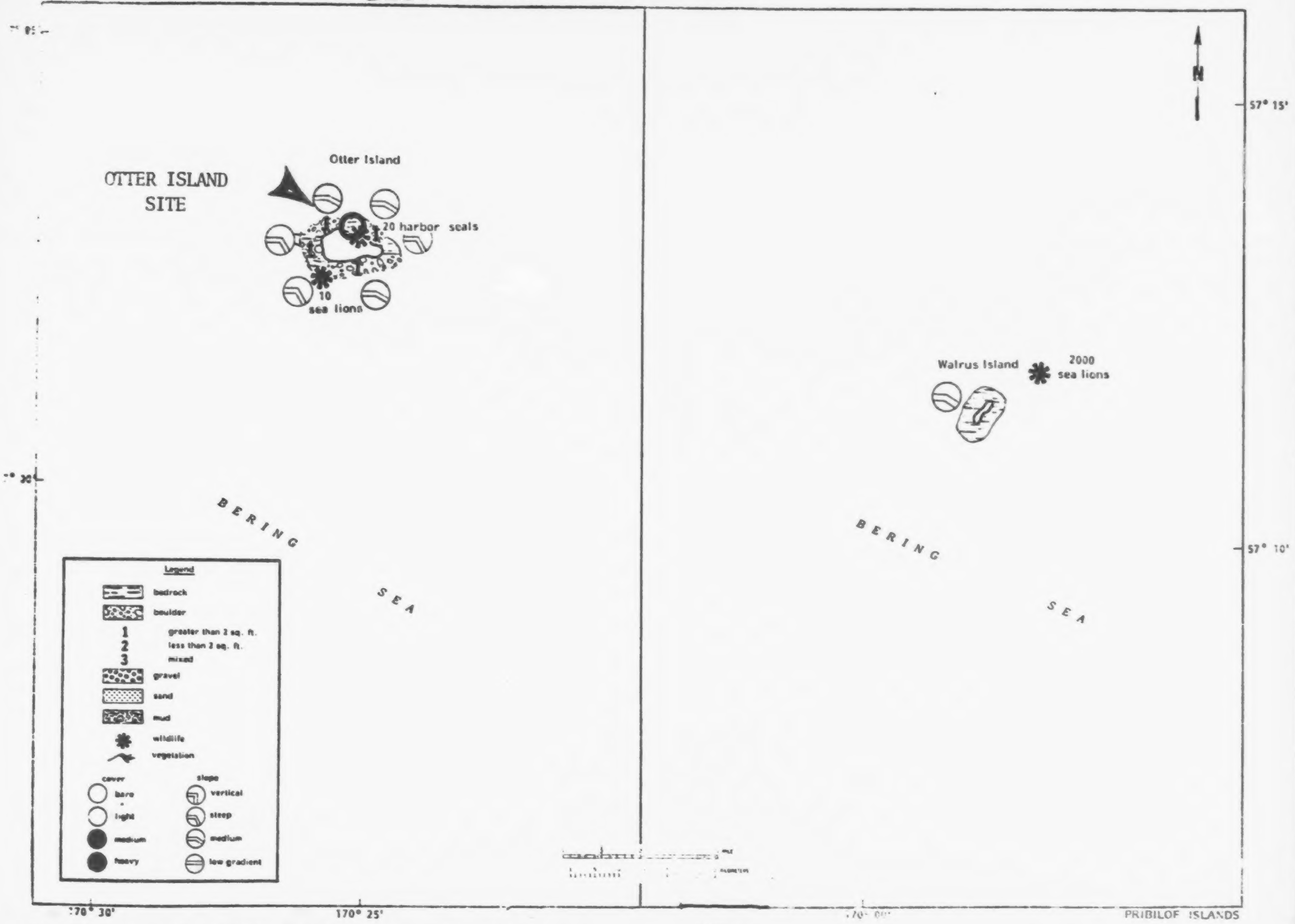
2.2 Otter Island

Otter Island, south of St. Paul Island (Figs. 43 and 45) resembles the profile of a sea otter floating on its back. The island has low hills at both ends (85.3 m and 45.7 m at the south and north ends respectively) and is low in the middle. This resemblance is reportedly the source of its name. The abrupt bluff on the south end of the island supports rookeries of auklets, murres, and kittiwakes (see Hunt 1977). A portion of the north shore of the island has a low gradient, and hair seals and Steller sea lions haul out there. Foxes are common and were seen in both 1975 and 1976 foraging for carrion in the intertidal zone.

The low gradient intertidal bench at the study site was characterized by volcanic rocks with many crevices of variable size separating numerous small hummocks. The hummocks exhibited many small pits and depressions in an otherwise smooth, polished surface. Table 13 summarizes the pertinent sampling information for Otter Island.

The upper intertidal zone (+0.6 to +2.4 m) contained much more bare rock than macrobiotic cover (Fig. 46). A film of algae (probably diatoms) made the rocks slippery. _Fucus_ was uncommon, being very squat but often fertile. _Halosaccion_, on the other hand, was found in luxuriant patches throughout much of the intertidal zone (+0.1 to +2.4 m). _Littorina sitkana_ was also found throughout this tidal range, but was usually confined to crevices at low tide. We observed it being preyed on by _Nucella lima_.

Mytilus edulis was present, although relatively few in number and small in size. They were limited almost exclusively to crevices, with a few large individuals found in one large crevice. _Modiolus modiolus_ and _Musculus discors_ were occasionally found with the small _Mytilus_.



OTTER ISLAND
SITE
Otter Island
20 harbor seals
10
sea lions
Walrus Island
2000
sea lions
BERING
SEA
BERING
SEA
N
57° 15'
57° 10'
170° 30'
170° 25'
PRIBILOF ISLANDS
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 45. Location of the study site at Otter Island.

Fig. 46. Upper half of Transect 1, Otter Island. 16 August 1975.

The lower intertidal zone (0.1 to 0.5 m; Figs. 47 and 48) was characterized by a canopy of the kelp *Alaria taeniata* and an understory of *Palmaria palmata*, *Iridaea cornucopiae*, *Pterosiphonia* sp., and filamentous diatoms. The rocks were often covered with patches of *Lithothamnion*. The snails *Margarites helicinus* and *Haloconcha reflexa* were seen grazing on the surface of the kelp, and the chiton *Schizoplax brandtii* was common on rocks. The crevices in this zone were filled with a diverse fauna including anemones, very dense aggregations of polychaetes (e.g. *Amphiglena pacifica*), gammarids (*Parallorchestes ochotensis*), and holothurians (*Cucumaria pseudocurata*).

Species richness in major taxonomic groups of organisms and species-abundance patterns among mollusks at Otter Island are given in a later section on the effects of ice scouring on intertidal communities.

2.3 St. George Island

2.3.1 Garden Cove--The two Garden Cove sampling sites were on an irregular coastline facing southeast on St. George Island (Figs. 43 and 49)--one on a rocky beach and the other on an adjacent sand beach (Fig. 50). Two parallel transects were placed 6 m apart on the rocky beach (Figs. 51 and 52; Table 13). One (T1) was sampled at even meters, the other (T2) was sampled every meter. In addition, eight 1/16 m^2 selectively placed quadrats were sampled (Table 13). Southwest of the survey site we made observations at an irregular rock outcrop somewhat protected by large offshore rocks from the effects of ice scouring.

Very little macroscopic life was seen on the transects probably because of ice scouring or disturbance by wave-induced boulder movement. A few littorines and an occasional amphipod were observed. Some rocks were entirely covered by filamentous algae and others were covered by diatoms.

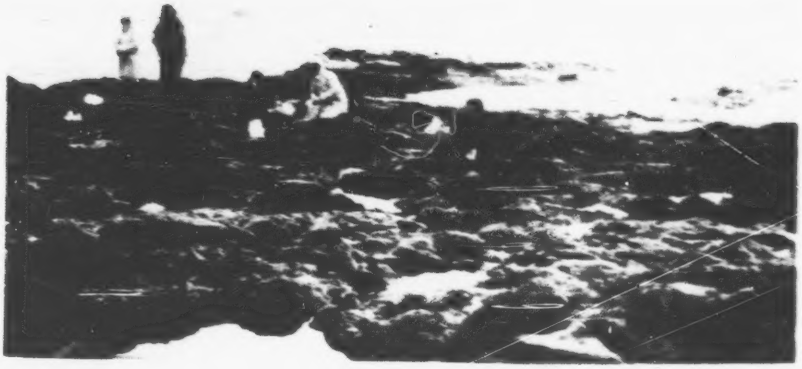


Fig. 47. Lower end of Transect 1-75 at Otter Island. 16 August 1975.

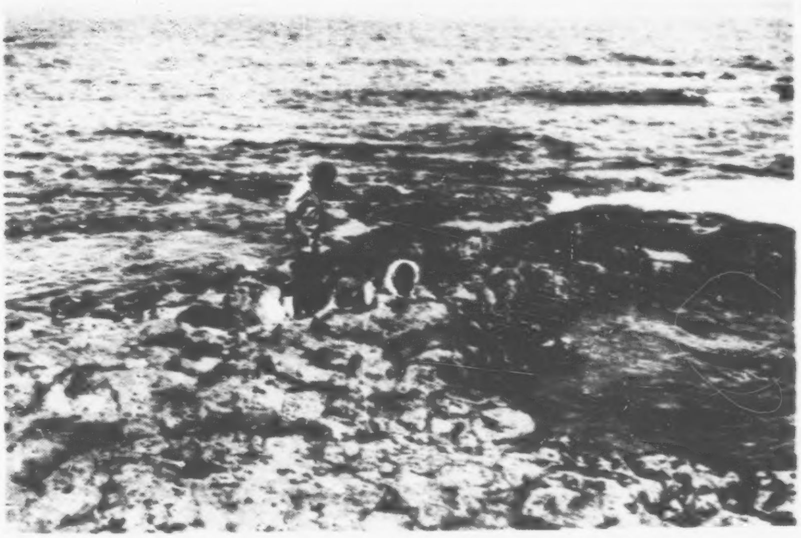
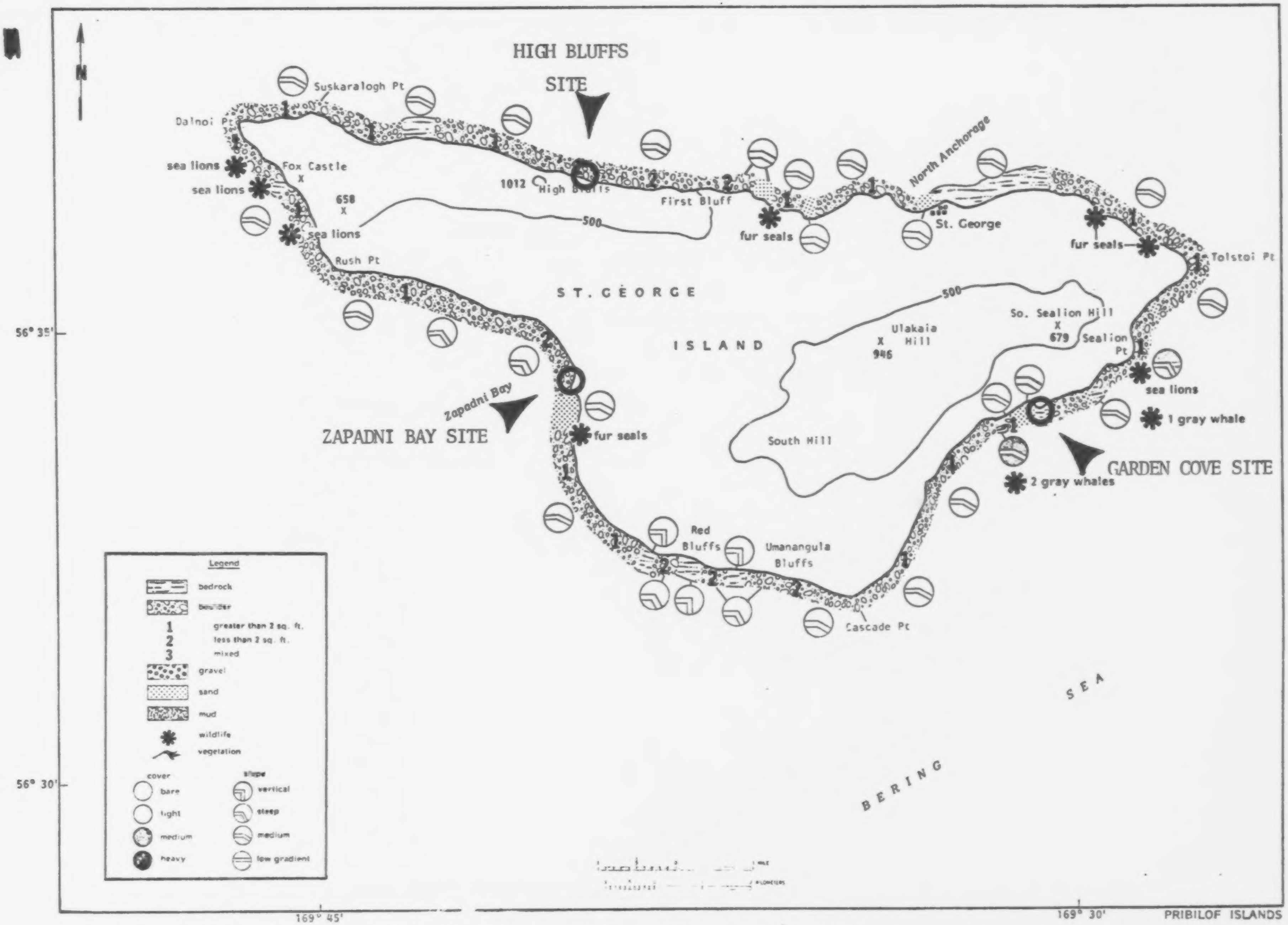


Fig. 48. Lower end of Transect 1-76 at Otter Island. 12 June 1976.



N
HIGH BLUFFS SITE
Suskaralogh Pt
Dalnoi Pt
sea lions
Fox Castle
sea lions
658
sea lions
Rush Pt
1012
High Bluffs
500
First Bluff
fur seals
North Anchorage
St. George
fur seals
Tolstoi Pt
ST. GEORGE
ISLAND
56° 35'
Zapadni Bay
ZAPADNI BAY SITE
fur seals
500
Ulakaia Hill
946
So. Sealion Hill
679
Sealion Pt
sea lions
1 gray whale
South Hill
GARDEN COVE SITE
2 gray whales
Red Bluffs
Umanangula Bluffs
Cascade Pt
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
56° 30'
BERING SEA
169° 45'
169° 30'
PRIBILOF ISLANDS

Fig. 49. Location of the three St. George Island sites: Zapadni Bay, High Bluffs, and Garden Cove.

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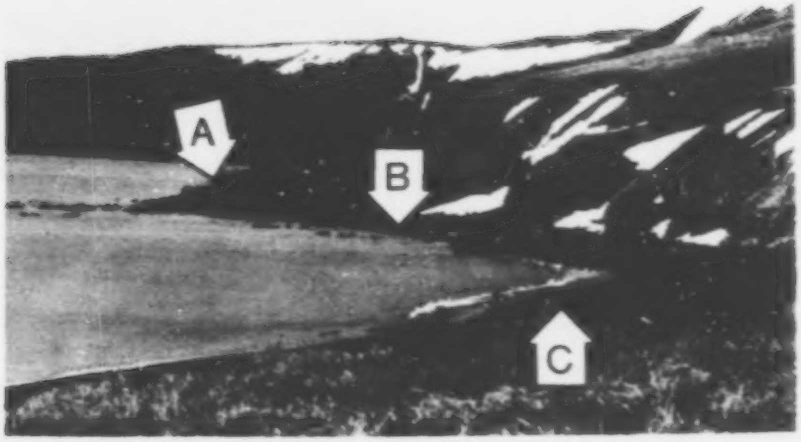

Fig. 50. Study sites at Garden Cove, St. George Island: A = rock outcrop protected from ice scouring; B = rocky beach transect area; C = sand beach transect area. 9 June 1976.

Fig. 51. Transect sites at Garden Cove, St. George Island. 9 June 1976.

Fig. 52. Lower end of Transect 1 at Garden Cove, St. George Island. 9 June 1976.

Halosaccion, Spongomorpha, Fucus, and an unidentified red alga were noted on a few rocks.

Many more species were observed in crevices at the rock outcrop site west of the transects. The species found included hydroids, ectoprocts, two kinds of encrusting sponges, urchins, nemerteans, and annelids including tube-dwelling polychaetes which surrounded very large Balanus cariosus. We also observed scale worms, caprellids, very large Collisella pelta, a large unidentified gastropod, Littorina, Nucella, Mopalia, two kinds of tunicates, and coralline algae.

Four 1-liter cores were taken at arbitrarily selected spots on the sand beach (Table 13). The cores contained very few macroscopic organisms including only Urospora sp., algae fragments, unidentified flatworms, oligochaetes, and cumaceans (Family Nannastacidae). Beached drift in the cove was sparse except at the eastern end of the beach where windrows of Laminaria and Alaria fistulosa were observed.

2.3.2 High Bluffs--The High Bluffs sampling site was on an exposed boulder beach on the north shore of St. George Island (Figs. 43 and 49). The study area was situated at the base of a high unstable cliff rising 200 to 300 vertical meters and occupied by large numbers of nesting birds (see Hickey and Craighead 1977, Hunt 1977). Rocks and pebbles frequently fell from the nearly vertical face and made the area a dangerous one in which to work. Many dead sea birds with massive injuries were found at the base of the cliff, presumably killed by falling rocks. The intertidal survey site was located at about the middle point of the High Bluffs area.

The beach at the sample site was relatively steep (slope, 30-40°) and consisted of large boulders in the high intertidal area grading into large angular blocks of stone with occasional boulders in the lower intertidal area.

The spaces between the boulders contained rubble and gravel. Many boulders had a polished appearance.

The sampling area appeared to be disturbed by ice scouring. Very few species were present. Littorines were abundant in crevices and common on outer surfaces of bare rock in the upper intertidal area. They were actively grazing the microalgal film covering the boulders. In the lower intertidal area, diatoms and green algae (*Spongomorpha* and *Urospora*) covered the seaward faces of the largest rocks. Large patches of *Halosaccion* (up to 83 g per 625 cm^2, wet weight) occurred on some boulders. *Iridaea* and *Fucus* were also noted. A few very small annelids or nematodes and ectoprocts were observed in crevices in the lower intertidal area.

2.3.3 Zapadni Bay--Zapadni Bay is located on the southwestern side of St. George Island (Figs. 43 and 49). Our intertidal station there was composed of a 10 m long boulder pile, 0.0 to +1.5 m above MLLW (Fig. 53). The upper portion of the intertidal area had relatively sparse biotic cover while the lower part (Fig. 54) was comparatively lush. Dates and methods of sampling at Zapadni Bay are summarized in Table 13.

The boulders in the upper intertidal zone had a patchy cover of a few large organisms and were covered with benthic diatoms that were being grazed by many *Littorina sitkana*. *Halosaccion glandiforme* and *Fucus distichus* occurred in dense patches on the tops and sides of some boulders. The *Fucus* had an unusual appearance at this station: both fertile and nonfertile plants appeared robust, but were squat; and all *Fucus* plants were less than 5-8 cm tall.

Between and beneath the boulders at the lower end of the pile were some animals whose abundance, because of their cryptic nature and our sampling methods, was not reflected in our quantitative collections. These included

Fig. 53. Upper end of systematic transect at Zapadni Bay, St. George Island. 15 August 1975.

Fig. 54. Lower half of systematic transect at Zapadni Bay, St. George Island. 15 August 1975.

many brooding anemones, dorid nudibranchs (feeding on the sponge *Halichondria panicea*), and unidentified compound ascidians.

The bedrock bench in the lower intertidal zone (-0.3 to 0.0 m) was generally flat but exhibited much small-scale relief (Fig. 54) with numerous tide pools and many small hummocks. Adjacent hummocks were often separated by crevices filled with coarse sand and broken shell.

The kelp *Alaria taeniata* formed a canopy in the lower intertidal zone. These plants occurred mainly with their holdfasts on the sides of the hummocks or in crevices. The holdfasts and crevices trapped sand and broken shell, making a microhabitat rich in organisms. These organisms included the sponge *Halichondria panicea*, nemerteans, many oligochaetes, polychaetes (*Chone gracilis*, *Fabricia sabella*, *Boccardia* spp., and *Exogone* spp.), *Ischyrocerus* sp., a few harpacticoid copepods, and bivalves (*Hiatella arctica*, *Protothaca staminea*, and small *Mytilus edulis*). Also present were *Cucumaria pseudocurata*, *Strongylocentrotus droebachiensis*, and the tunicate *Molgula oregonia*. *Laminaria groenlandica* occurred with the *Alaria* but was not as abundant. Commonly found foraging on both kelps were the gastropods *Haloconcha reflexa* and *Margarites helicinus*. Egg masses of *Margarites* were quite common on the kelp blades.

A few chitons including *Katharina tunicata* and *Schizoplax brandtii*, and the gastropods *Buccinum baeri* and *Nucella lima* were found. Many large (2-4 cm) *Collisella pelta* were seen. No small (<1 cm) limpets were observed. The hummocks were often covered with patches of *Lithothamnion* and *Ralfsia*. Other algal species here included *Iridaea* sp., *Palmaria palmata*, and occasional tufts of *Pterosiphonia* and *Ptilota*.

The barnacles *Balanus glandula*, *B. balanoides*, *B. cariosus*, and *Chthamalus dalli* were conspicuously low in abundance. Although *B. cariosus* did occur occasionally on the transect, extensive patches of this and other species of barnacles were absent. Because the Pribilof Islands are near the southern limit of Bering Sea ice, the presence of sea ice in some years may preclude the development of extensive barnacle populations, either physically by scouring or physiologically by hampering growth and development. The presence of sea ice in winter and early spring in some years may also help to explain the squat habitus of the Fucus.

A few deep (20 cm) tide pools were among the hummocks. Often these tide pools had numerous anemones in them, and much of their perimeter was inhabited by the sponge Halichondria panicea. Occasionally caprellids and the gastropod Velutina plicatilis could be seen on the sponge. Numerous pycnogonids (predominantly Ammothea pribilofensis and Achelia chelata) occupied the bottoms of the pools. Considerable size variation existed among the pycnogonids. A few were ovigerous males. More information on the frequency of occurrence and biomass of dominant organisms at Zapadni Bay is included in a later section on the effects of ice scouring on intertidal communities.

3. Norton Sound

We sampled intertidal communities at 13 locations in or near Norton Sound (Fig. 55) once in August 1976. Over half (54%) of the sites sampled were predominantly sandy beach; one fifth (23%) had a bedrock or boulder/bedrock substrate; and the remainder were beaches of gravel, mud or peat (Table 14). Zimmerman et al. (1977) show the relative percentages of the major intertidal substrates along the coast of Alaska from Sheldon's Point on the Yukon River delta north to Cape Prince of Wales. Table 14 shows the geographic coordinates, sampling date, and characteristics of the beach and surface seawater at each site.

The tidal range in Norton Sound is relatively narrow: the diurnal range (the difference in height between MHHW and MLLW) ranges from 128 cm at the Pikmiktalik River entrance in the south to 49 cm at Nome (National Oceanic and Atmospheric Administration, 1976). The predicted heights of low water during our visit to Norton Sound ranged from 14 cm below MLLW to 11 cm above MLLW.

Rocky shores were sampled with a transect along which 1/16 cm^2 quadrats were collected at meter intervals (Table 15). Sand, gravel, and peat beaches were usually divided into three zones (high, middle, and low) at arbitrarily designated tidal heights; two to four samples were taken from each zone. The sampling locations were determined haphazardly. Though not strictly random, this method of determining sampling locations was probably not seriously biased because usually the substrate was superficially homogeneous and most organisms were infaunal; therefore, obvious visual cues which might bias an investigator were absent.

All gravel, sand, or mud samples were sieved on site through one or two sieves, the finest having 2 mm openings. Because of the large mesh openings

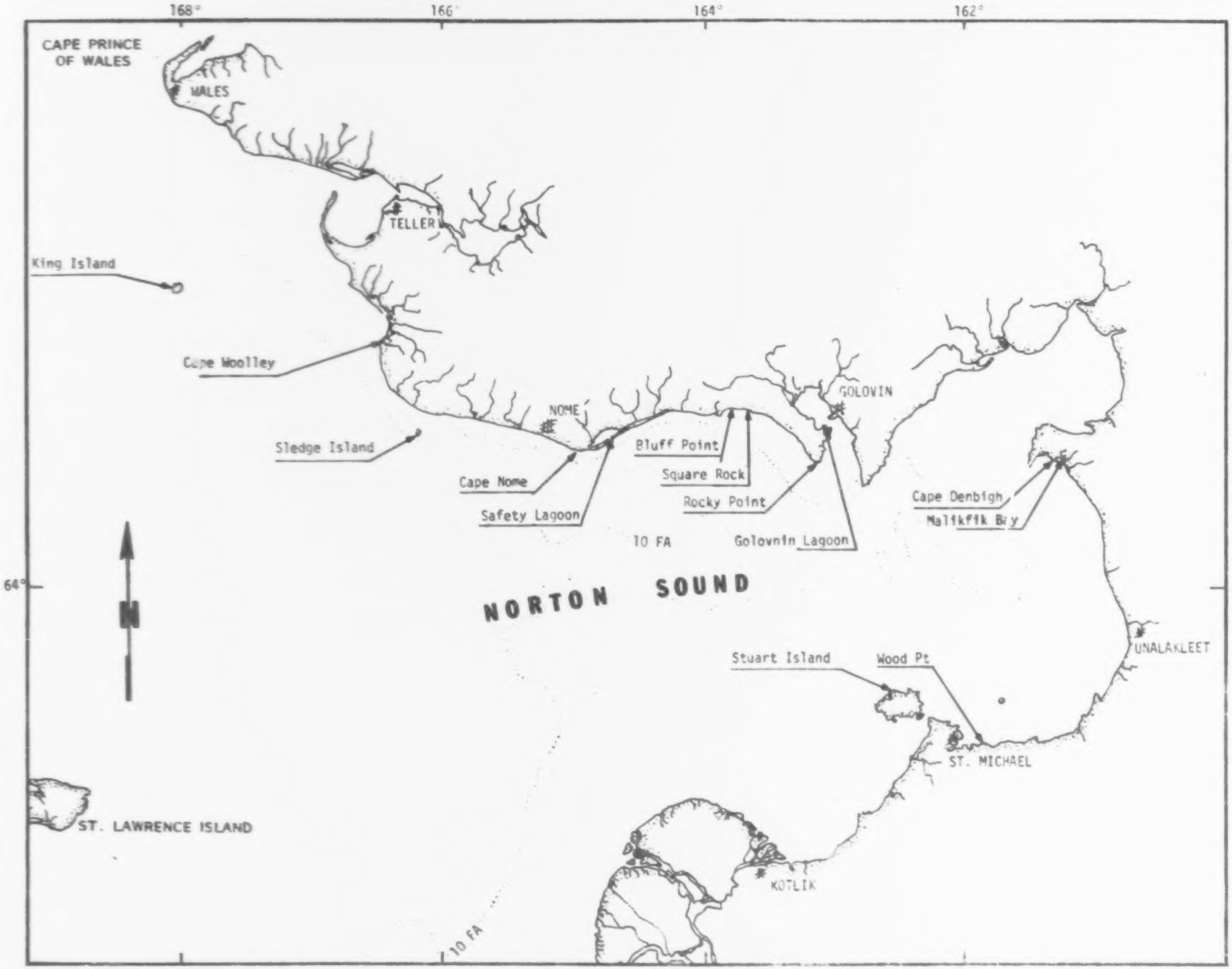
168°
166°
164°
162°
64°
CAPE PRINCE OF WALES
WALES
TELLER
King Island
Cape Woolley
Sledge Island
NOME
Cape Nome
Safety Lagoon
Bluff Point
Square Rock
Rocky Point
Golovnin Lagoon
GOLOVIN
Cape Denbigh
Malikfik Bay
10 FA
NORTON SOUND
N
UNALAKLEET
Stuart Island
Wood Pt
ST. MICHAEL
ST. LAWRENCE ISLAND
KOTLIK
10 FA

Fig. 55. Location of sites in Norton Sound.

Table 14. Geographic coordinates, sampling dates, and beach- and surface-seawater characteristics at 13 localities in Norton Sound where intertidal communities were sampled.

Site	Latitude (N)	Longitude (W)	Date (Aug 76)	Beach Characteristics Substrate	Slope (°)	Orientation (°)[a]	Surface-Seawater Characteristics Temperature (°C)	Salinity (°/oo)
Stuart Island	63°38'	162°33.5'	8	bedrock	5	000	13.3	19
Wood Point	63°27.4'	161°45'	9	boulder/bedrock	--	--	--	--
Wood Point	63°27.4'	161°45'	9	peat	20	340	15.6	18
Malikfik Bay	64°23.8'	161°16.2'	10	mud/peat	0	20	20	18
Cape Denbigh	64°23.9'	161°19'	10	sand	10	180	--	--
Golovnin Lagoon	64°30.6'	163°05.9	11	sand	25	310	--	--
Rocky Point	64°24.5'	163°10.7'	11	sand	10-15	250	13.3	21.5
Square Rock	64°33.6'	163°34.2'	12	gravel	10-15	180	15.3	19.5
Bluff	64°34.6'	163°49.9'	12	gravel/sand	25	190	--	--
Safety Sound	64°28.4'	164°42.6'	14	sand/mud	<3	320	13.3	21.4
Cape Nome	64°28.6'	164°43.3'	14	sand	<5	--	--	--
Sledge Island	64°28.9'	166°12.5'	7	sand	--	--	9.7	27.1
Cape Woolley	64°50.2'	166°23.8'	13	sand/gravel	20	240	12.8	21.3
Cape Woolley	64°47.9'	166°28.1'	13	bedrock	--	300	--	--
King Island	64°57.7'	168°03.9'	6	boulder	--	--	--	--

a. Bearing with reference to magnetic North.

Table 15. Pertinent sampling information for 14 intertidal sites in Norton Sound.

Site	Tidal Range Sampled(cm)	Sampling Methods	Sample Surface (cm^2) or Volume(ℓ)[a]	Sample Size (n)
Stuart Island	+3 to +85	Transect	625	14
Wood Point	not recorded	Haphazard	1	10
	+198 to +286	Transect	625	16
Malikfik Bay	+15 to +58	Haphazard	7	15
Cape Denbigh	+6 to +119	Selected	7	8
Golovnin Lagoon	+6 to +94	Haphazard/Selected	7,1	9,4
Rocky Point	+6 to +119	Haphazard	8	6
Square Rock	+15 to +128	Haphazard	8	5
Bluff	+40 to +192	Haphazard	7	6
Safety Sound	-46 to -24	Haphazard	8	8
Cape Nome	+3 to +85	Haphazard	7,1	6,2
Sledge Island	+18 to +158	Selected	3.5	6
Cape Woolley	+18 to +140	Haphazard	7	8
King Island	not recorded	Qualitative	--	--

a. Surface area of quadrat at Stuart Island and Wood Point Transects; volume at other sites.

of the sieves, only the larger macroscopic organisms were adequately sampled. Abundances of Nematoda, Oligochaeta, Cumacea, Mysidacea, and most species of Polychaeta, Gammaridea, and larval Insecta were probably underestimated because small individuals passed through the sieve.

Species richness at our rocky intertidal sites (Stuart Island and Wood Point; Table 16) was similar to that on ice-stressed shores in the Pribilof Islands (see later section on effects of scouring by sea ice on intertidal community structure). Fucus distichus had the greatest biomass of all organisms and Chthamalus dalli was the most abundant invertebrate at our rocky sites.

Comparisons of species richness among sites with unconsolidated substrates were difficult to make because sample volume varied among sites. When all samples were corrected to 8-liter, sites with a peat or peat and mud substrate showed the greatest species richness (Table 16), but this may have been an artifact resulting from the extrapolation of 1-liter samples to 8 liters. The most commonly occurring organisms on the unconsolidated beaches were filamentous blue-green algae, polychaetes, gammarids, and insects. All species with live representatives at our Norton Sound sites are listed in Appendix II-D.

3.1 Stuart Island

Stuart Island is in southeastern Norton Sound (Fig. 55). Our study site was on a gently-sloping rock bench on the northern side of the island (Fig. 56). The substrate consisted of igneous rock fractured into blocks and some loose boulders (Fig. 57). In addition to the samples that were taken on the transect (Table 15), a single selected sample was taken nearby.

The abundance of algae and invertebrates in quadrats on the transect is shown in Table 17. Two quadrats contained only unrecorded amounts of

Table 16. Average species counts of plants and invertebrates[a] in quadrats or cores at intertidal sites in Norton Sound.

Site	Sample Area (cm^2) or Volume(ℓ)[b]	Upper Intertidal Area			Lower Intertidal Area		
		N	$\bar{x}$[c]	S	N	$\bar{x}$[c]	S
Stuart Island	625	11	4.6	2.5	4	9.8	1.7
Wood Point							
peat	1	9	2.5(20)	1.9	-	-	-
boulder/bedrock	625	13	6.8	3.3	-	-	-
Malikfik Lagoon	1	10	2.1(16.8)	1.8	5	1.4(11.2)	0.6
Cape Denbigh	7	6	1.2(1.4)	1.5	2	1.5(1.7)	0.7
Golovnin Lagoon	7	6	1.3(1.5)	0.8	3	0.3(0.3)	0.6
Rocky Point	8	5	1	1.7	1	3	-
Square Rock	8	4	1.5	1.3	-	-	-
Bluff	7	6	1.2(1.4)	1	-	-	-
Safety Sound	8	1	0	-	7	2.9	1.8
Cape Nome	7	2	2(2.3)	2.8	4	0.2(0.2)	0.5
Sledge Island	3.5	4	0.5(1.1)	1	2	1.5(3.4)	0.7
Cape Woolley	7	5	0.8(0.9)	0.8	1	0	-

a. Includes all organisms except those belonging to the following taxa: Porifera, Cnidaria, Platyhelminthes, Nemertea, Oligochaeta, Copepoda, Tanaidacea, Insecta (except *Anurida maritima*), Arachnida, Acarina, Sipuncula, Ectoprocta, and Ascidiacea.

b. Surface area of quadrat at Stuart Island and Wood Point, boulder/bedrock; volume at other sites.

c. Numbers in parentheses are means standardized to 8 liters.

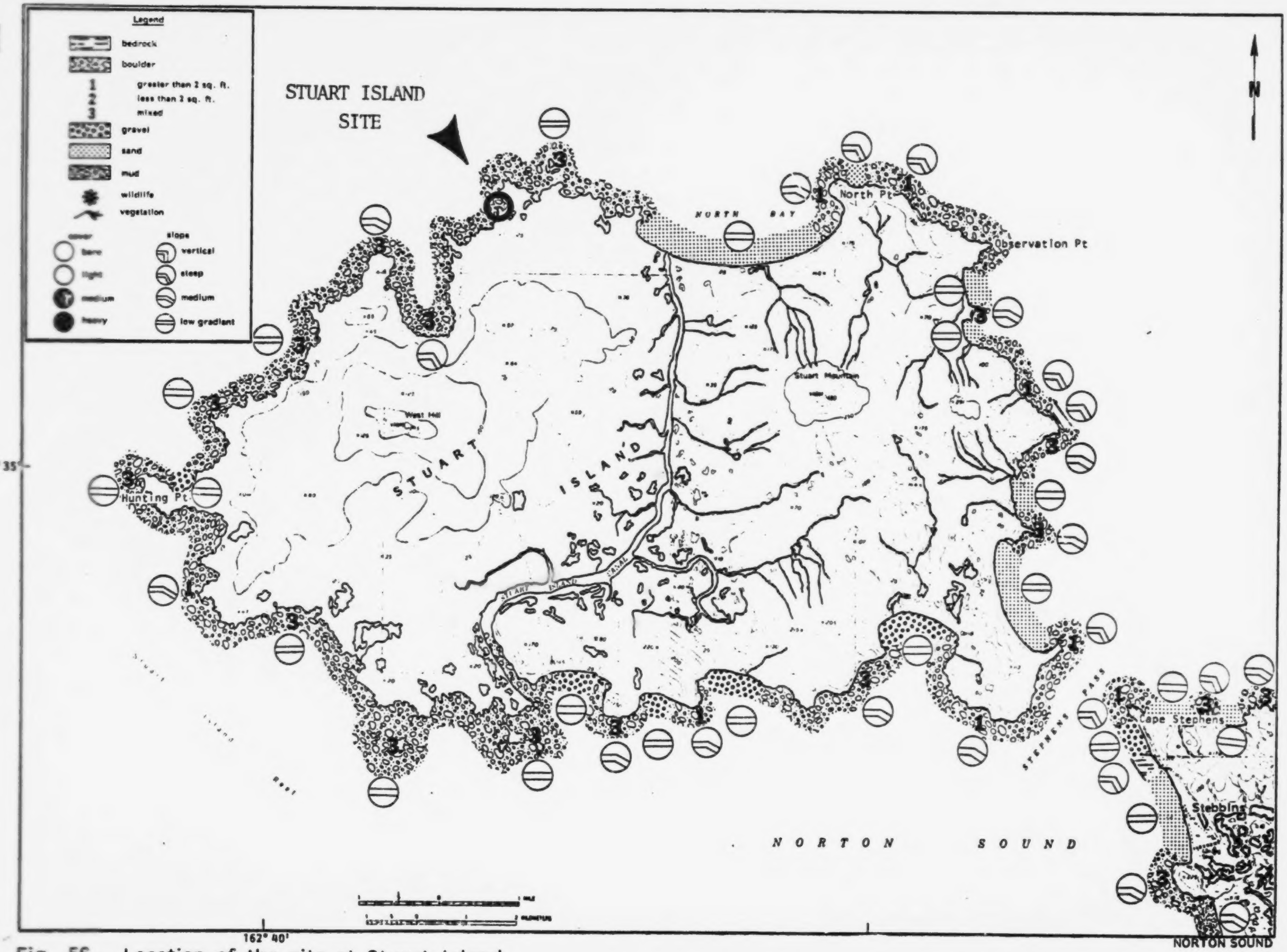
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
STUART ISLAND SITE
N
NORTH BAY
North Pt
Observation Pt
Stuart Mountain
West Hill
STUART ISLAND
Hunting Pt
63° 35'
STEPHENS PASS
Cape Stephens
Stebbins
NORTON SOUND
162° 40'
NORTON SOUND

Fig. 56. Location of the site at Stuart Island.

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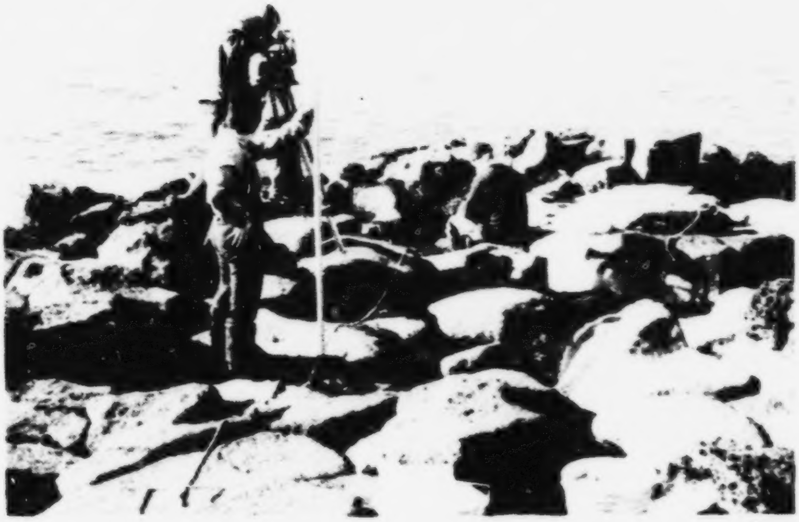

Fig. 57. The transect at Stuart Island, Alaska. 8 August 1976.

Table 17. Abundances of species of algae and invertebrates on the transect at Stuart Island, Norton Sound.

	Abundance[a]												
Sample No.	1-5	1-2	1-6	1-3	1-4	1-7	1-9	1-8	1-12	1-13	1-10	1-11	1-14
Species / Elevation (cm)	85	76	70	67	64	55	52	46	46	37	30	24	3
Monostroma sp.					0.2								
Enteromorpha clathrata		20+		6.0	2.3				0.2				
E. intestinalis								<0.1					
E. linza										6.8[b]			
Ulva lactuca				<0.1			0.9		3.7	2.7	28.4	1.2	8.7
Lola lubrica											<0.1		
Spongomorpha cf *S. saxitilis*													88.4
Pylaiella littoralis			4.7						7.7	3.8	1.6		
Melanosiphon intestinalis													2.4
Petalonia fascia										3.2		<0.1	
Scytosiphon sp.										9.9		4	
Fucus distichus		102.8	47.2	15.9	275.0	216	577.9	467.3	293.9	133.4	61.8	129.8	114.3
Gigartina pacifica													32.5
Pterosiphonia bipinnata					<0.1								
Acrochaetium sp.							38.5						
Eteone longa					1								
Spio filicornis												1	
Fabricia sabella		56	2										
Lacuna sp.											8	3	
L. vincta											1	13	0.1
L. variegata									8				
Littorina sitkana									2			1	
L. squalida													
Mytilus edulis							1				1	3	
Chthamalus dalli	210	663	820	70	316	435	489	702	544	112	702	1438	2
Gammarus sp.			33	14	19				70		10		
Anisogammarus sp.							38			4	11	85	149

a. Number of individuals of animals, wet weight (g) of plants.

b. Sample contained some *E. clathrata*.

unidentified barnacles and are not included in the analysis. Organisms collected in the plots, but not shown in Table 17 included fragments of algae; mites; oligochaetes; nematodes; and chironomid, dipteran, and coleopteran larvae.

Fucus distichus had greatest biomass in most quadrats (Table 18). *Chthamalus dalli* occurred in all quadrats, but its wet weight exceeded that of *Fucus* in one quadrat only. *Fucus* and *Chthamalus* together made up over 50% of the biomass in all quadrats, except one in which *Spongomorpha* cf. *S. saxitilis* replaced *Chthamalus* as a codominant with *Fucus* (Table 17). The remaining species of algae that averaged relatively high amounts of biomass were primarily ephemeral or fugitive species (Table 18; see section on effects of ice scouring for a discussion of ephemeral and fugitive algae).

Among the other invertebrates at our Stuart Island site, gammarid amphipods and the sabellid polychaete *Fabricia sabella* were patchily abundant. Herbivorous gastropods such as *Lacuna* spp. and *Littorina* spp. were uncommon. *Mytilus edulis* was small and rare.

3.2 Wood Point

Wood Point is on the southeastern shore of Norton Sound east of Stuart and St. Michael Islands (Figs. 55 and 58). Our study site was composed of two types of substrate, peat and boulder-bedrock, which we sampled with separate transects. The substrate at the peat site consisted of dense, organic mud with large igneous rocks scattered over the surface. The substrate at the rocky site consisted primarily of large angular boulders (Figs. 59 and 60).

At the peat site, nine 1-liter cores were taken: three each at high-, middle-, and low-tidal levels. A surface sample and a deep core were taken in the lowest tidal zone. At the rocky site, we collected quadrats at meter

Table 18. Statistics for 10 species of algae and invertebrates which had the greatest average rank in quadrats at Stuart Island, Norton Sound.

Species	Average Rank[a]	Wet Weight(g[b])	Frequency of Occurrence[c]	Abundance[d]	Dominance[e]
Fucus distichus	6.1	131.6, 15.9-577.9	0.92	---	11/13
Chthamalus dalli	4.8	26.1, 0.07-98.2	1.0	489, 2-1428	4/13
Ulva lactuca	2.8	2.1, 0.04-28.4	0.62	---	0
Pylaiella littoralis	1.9	3.8, 0.03-7.7	0.38	---	0
Anisogammarus sp.	1.5	0.6, 0.05-1.6	0.38	38, 4-149	0
Scytosiphon sp.	1.3	7, 4-9.9	0.15	---	0
Enteromorpha clathrata	1.2	4.3, 0.2-20+	0.31	---	0
Gammarus sp.	1.2	0.3, 0.06-1.2	0.38	33, 10-70	0
Lacuna vincta	0.8	0.08, 0.02-0.2	0.23	6, 1-13	0
Lacuna sp.	0.7	0.08, 0.06-0.1	0.15	6, 3-8	0

a. Rank of wet weight of each species averaged over 13 quadrats (1, lowest weight). Species listed in order of decreasing rank.

b. Median and range of wet weights per 1/16 m^2 in all quadrats where the species was found.

c. Proportion of quadrats that contained the species.

d. Animals only, median and range of number of individuals per 1/16 m^2 in all quadrats where the species was found.

e. Proportion of quadrats in which the species was among those making up 50% of the wet weight. Summation in each quadrat began with the heaviest species.

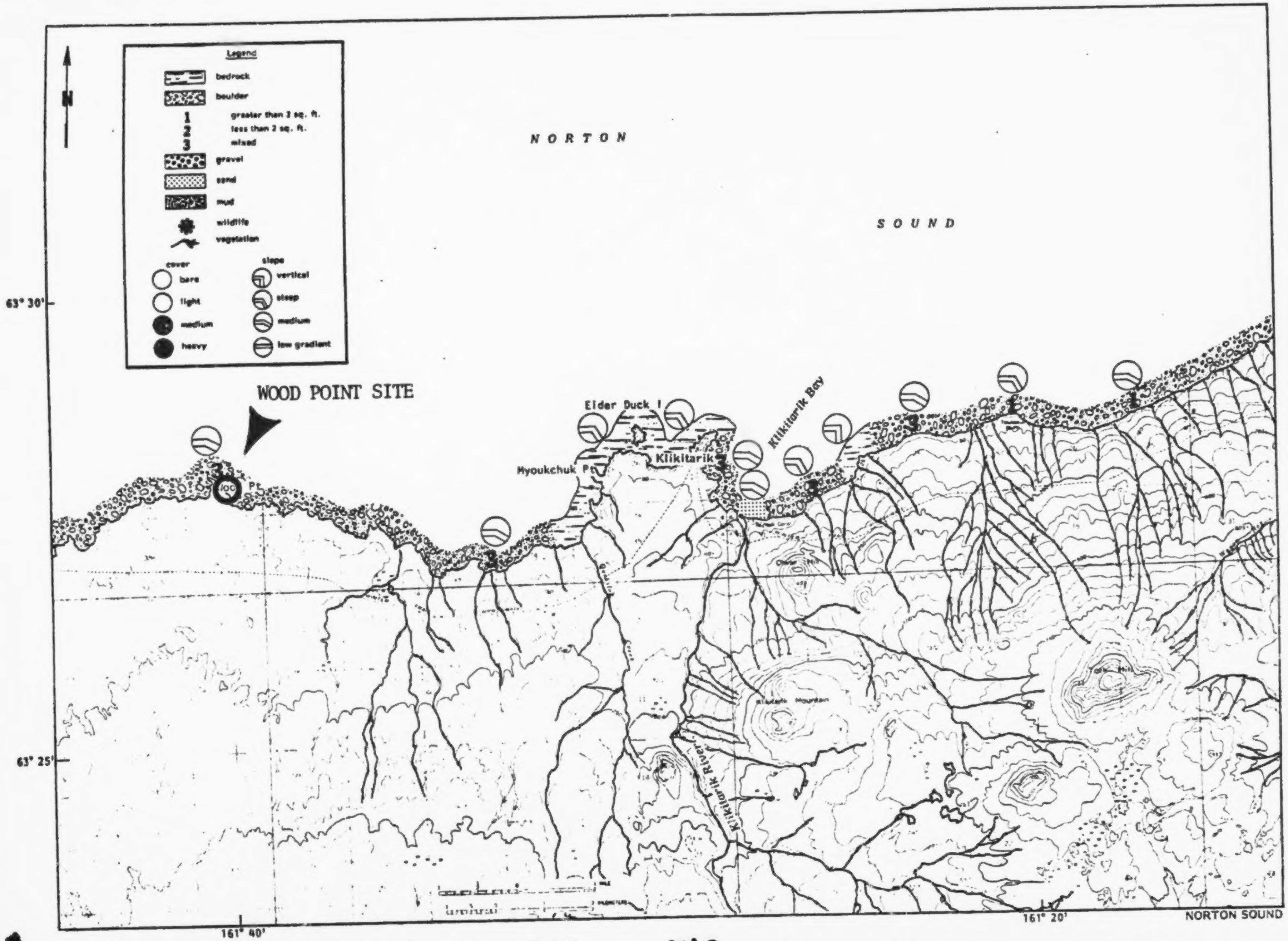
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
N
NORTON
SOUND
63° 30'
63° 25'
161° 40'
161° 20'
WOOD POINT SITE
Wood Pt
Elder Duck I
Myoukchuk Pt
Klikitarik
Klikitarik Bay
Klikitarik River
NORTON SOUND

Fig. 58. Location of the study area at Wood Point.

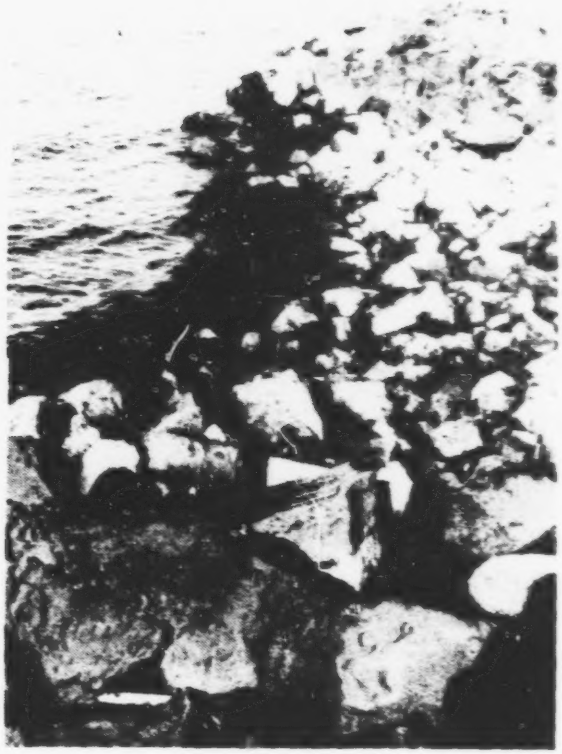

Fig. 59. Lower end of transect at Wood Point. 9 August 1976.

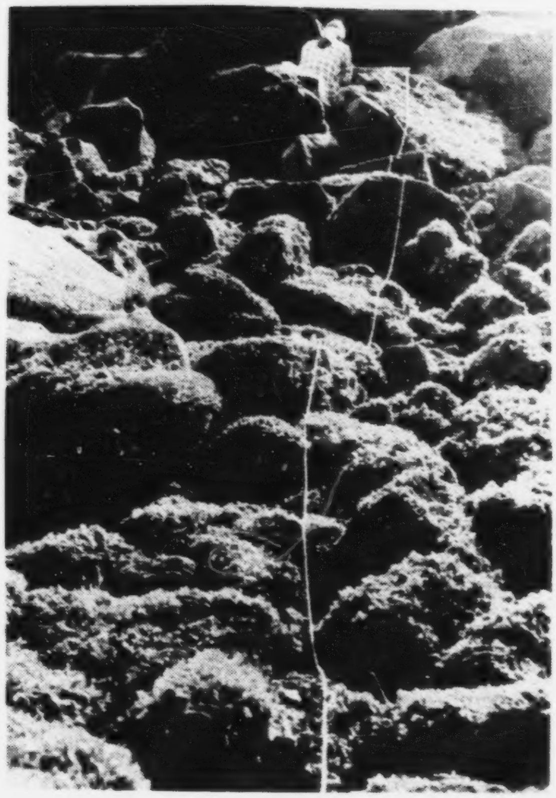

Fig. 60. Upper end of transect at Wood Point. 9 August 1976.

intervals along the transect (Table 19). One additional quadrat was collected at 10 m on the transect.

The abundances of most distinquishable species of algae and invertebrates collected in the cores at the peat site are shown in Table 20. Two cores contained no macroscopic organisms. One contained only unidentifiable algal fragments. Other biota in the cores included colonial diatoms, and fragments of algae, seagrass, and ectoprocts. Most of the species found in the cores are characteristic of rocky shores and may have drifted onto the mud from the adjacent boulder beach. The biomass of even the largest species in the cores was low (Table 20). One bucketful of mud collected near the low-tide contour and sieved through a 2 mm sieve contained only Anisogammarus sp.

3.3 Malikfik Bay

Malikfik Bay is a tidal flat of peat and mud covered by terrestrial grasses and laced with tidal channels and shallow, brackish ponds (Figs. 55 and 61). The outer area facing Norton Sound is sand covered with deep ripple marks (Fig. 62). The lagoon is protected, and when we were there the water was warm and brackish. We collected fifteen 1-liter cores, five each in the low-, mid-, and upper-tidal zones (Table 21, Fig. 63). The quantitative samples yielded very small numbers of plants and animals. Algae were represented by three species of blue-green algae (Cyanophyceae). Lewis (1964, p. 163) has observed that blue-green algae are abundant in salt-marsh turf-habitat in Scotland. Polychaetes were more numerous than other groups, constituting about one-third of the animal species. Of greater interest was the presence of the shrimp Crangon sp., not often noted in intertidal sampling, especially in mud cores such as we used.

Table 19. Abundances of the most frequently occurring species of algae and invertebrates on the rocky intertidal transect at Wood Point.

	Abundance[a]													
Sample No.	1-0	1-4	1-1	1-2	1-5	1-6	1-10	1-10A	1-7	1-9	1-8	1-13	1-11	1-12
Species Elevation(cm)	286	268	262	259	247	235	235	235	232	229	222	220	213	213
Enteromorpha clathrata	24.4		0.9				0.01		0.02b	0.04				
Pylaiella littoralis	15.6		2.5				0.01		0.01		0.4	0.2	0.01	
Sphacelaria racemosa			0.01									0.03	<0.01	
Fucus distichus	19.3	20.3	200.4	18.4	50.6	8.9	215.5	22.0	114	122.5	145.8	447.4	336	253
Ahnfeltia plicata	0.1		2.0	0.03	0.2		0.2		0.6	0.09	2.2	2.2	2.1	
Iridaea sp.				0.09										
Pterosiphonia bipinnata			0.2	0.07		0.04			0.04	0.01			0.24	
Pterosiphonia sp.					0.02		0.04				3.4	2.6		0.03
Abietinaria sp.												0.06	0.04	
Thuiaria sp.									<0.01			0.02		
Halecium sp.	0.02				<0.01		0.02		0.02				0.2	
Sphaerosyllis sp.			1											
Capitella capitata			2											
Mytilus edulis			2											
Musculus sp.												1		
Chthamalus dalli				20	16	22	2			2	2	14	18	57
Saduria entomon													1	
Gammarus sp.				1						12	2.1c	8		3
Anisogammarus sp.													5	
Eucratea loricata												0.02		

a. Wet weight (g) of plants, hydroids, and bryozoans, number of individuals of animals.

b. *Enteromorpha* cf. *E. clathrata*.

c. Wet weight (g); individuals not counted.

Table 20. Abundances of species of algae and invertebrates at the peat site at Wood Point, Norton Sound.

		Abundance[a]					
	Sample No.	BH-1	BM-1	CM-1	AL-1	BL-1	CL-1
Species	Elevation(cm)	296	--	--	204	204	204
Fucus sp.						0.3f[b]	
Fucus distichus					0.3f		0.2f
Acrochaetaceae			<0.01				
Ahnfeltia plicata		<0.01	0.06		<0.01f	0.04	
Gigartinaceae			<0.01		0.1f	0.04	
Iridaea sp.							0.1
Pterosiphonia bipinnata		0.06	0.04			0.01	0.1
Oligochaeta		52	2	23			
Gammaridea					1		

a. Number of individuals of animals, wet weight (g) of plants.
b. f = fragments.

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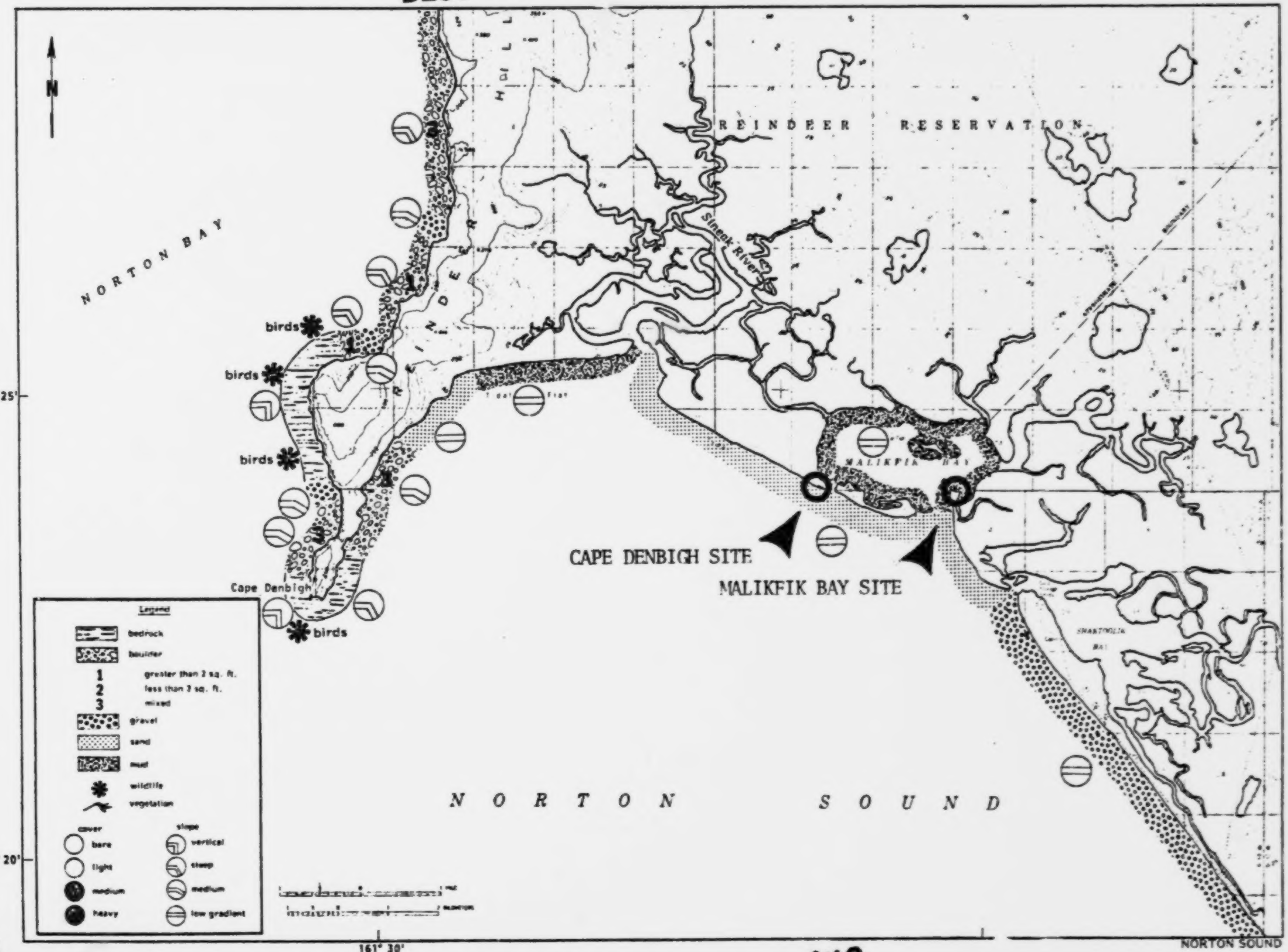
N
NORTON BAY
REINDEER RESERVATION
Sinéok River
birds
birds
birds
birds
Tidal Flat
MALIKFIK BAY
CAPE DENBIGH SITE
MALIKFIK BAY SITE
Cape Denbigh
SHAKTOOLIK BAY
NORTON SOUND
64° 25'
64° 20'
161° 30'
NORTON SOUND
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 61. Location of the Cape Denbigh and Malikfik Bay study sites.

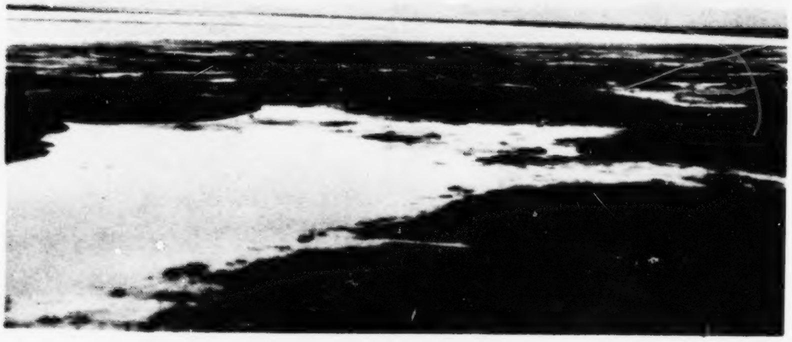

Fig. 62. Wetlands in the vicinity of Malikfik Bay. 10 August 1976.

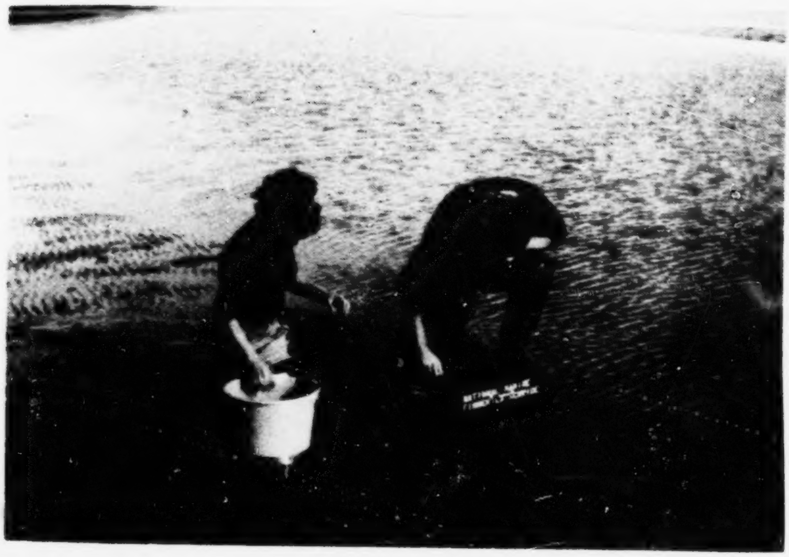

Fig. 63. Study area on a beach seaward of Malikfik Bay. 10 August 1976.

Table 21. Abundances of species of algae and invertebrates in 1-liter cores at Malikfik Bay, Norton Sound.

	Abundance[a]														
Plot No.	U-1	U-2	U-3	U-4	U-5	M-1	M-2	M-5	M-3	M-4	L-1	L-2	L-5	L-3	L-4
Species Elev.(cm)	58	58	58	55	52	40	40	40	37	37	18	18	18	15	15
Oscillatoriaceae	0.004			0.002				0.2							
Microcoleus/Lyngbya?		+	0.6		0.1							0.008		0.2	0.2
Nemertea								1							
Eteone longa						1	1								
Glycinde picta							1								
Pygospio elegans										4	3		.004		
Pygospio sp.							1	6	2						
Rhynchospio sp.												1			
Arenicola glacialis							1								
Oligochaeta			14			2		1	0.001						
Turtonia occidentalis						1									
Copepoda			1												
Saduria entomon	1						3	1							
Crangon sp.							1								
Insecta	1								1p[b]						
Chironomidae										1					

a. Number of individuals of animals, wet weight (g) of plants.
b. P = pupa.

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The tidal flat and the sand beach seaward of it were examined on foot and characterized. The shallow tidal channels contained numerous small flatfish, about 5 cm long, and several shrimp, 5 to 10 cm long. The channels covered a large area, and every channel contained many of these shrimp and small fish. This protected area could be a nursery supporting large populations of juvenile fish. The sand beach facing Norton Sound was littered with drilled shells of clams of several species (Fig. 64); the shells were probably drilled by moon snails (Family Naticidae), whose egg collars were abundant on the beach. We found no live molluscs, so we assume such populations are largely subtidal.

3.4 Cape Denbigh

Cape Denbigh is a rocky promontory that juts into Norton Sound at the southeastern corner of Norton Bay (Figs. 55 and 61). Our study area east of the Cape was a south-facing sand beach with a gentle 10° slope. The beach was divided into four tidal strata, and we dug two 2-liter samples from each stratum (Table 22).

Samples from the upper tidal stratum contained only empty bivalve shells (Macoma sp. and Clinocardium sp.) and sea-grass fragments. The only live organisms in the remainder of the samples were the polychaete Eteone longa and crustaceans. The lysianassid gammarid Onisimus littoralis occurred most frequently (present in 83% of our samples) and had the greatest average abundance (5.3 individuals/sample) at mid- and low-intertidal levels. The remains of several species of bivalves (Macoma sp., M. lama, M. balthica, Siliqua alta, Tellina lutea, and Mya sp.) and an unidentified gastropod were found in three samples from mid- and low-tidal levels. Many shells of these and other species of molluscs (i.e. Natica clausa, Polinices pallida, Serripes

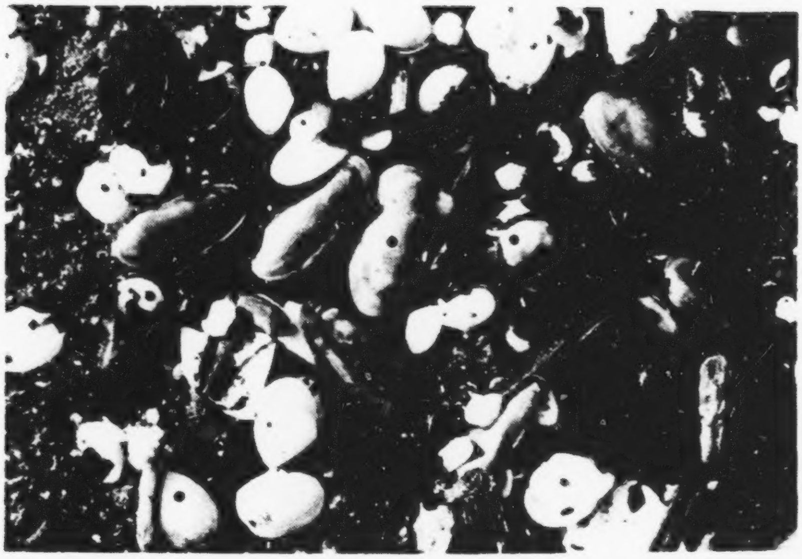

Fig. 64. Bivalve shells (probably Siliqua alta and other unidentified species) drilled by naticid snails. The clams were among the drift material on the beach seaward of Malikfik Bay. 10 August 1976.

Table 22. Abundances of invertebrates in sand samples from Cape Denbigh, Norton Sound.

				Abundance[a]			
	Sample No.	AM-1	BM-1	AN-1	BN-11	AL-1	BL-1
Species	Elevation (cm)	49	49	34	34	6	6
Eteone longa						1	1
Neomysis czerniawskii					25		
Pontoporeia affinis					2		
Onisimus littoralis		15	4	7	4	2	
Crangon sp.					1		

a. Number of individuals/quadrat.

groenlandicus, Clinocardium californiense and Solariella sp.) were scattered over the beach, presumably washed ashore from the sublittoral region.

3.5 Golovnin Lagoon

Golovnin Lagoon is located at the head of Golovnin Bay in northcentral Norton Sound (Figs. 55 and 65). We sampled a sand beach on the south shore of the Lagoon (Fig. 66). The site was chosen because we saw from the air an apparently dense eelgrass bed nearby; closer inspection revealed that the eelgrass bed was less dense than it had appeared.

The Lagoon was separated from a small bay to the southeast of it by a 1.5 km long sand spit. On the northwest side of the spit, we divided the beach into three zones based on tidal elevation. In each zone three 7-liter samples each comprising seven haphazardly placed 1-liter cores were collected (Table 15). In addition, a 1-liter core was taken in the adjacent eelgrass bed. On the southeast shore of the spit, three 1-liter cores were collected.

Sea-grass fragments (presumably eelgrass, Zostera marina) were present in all of the 7-liter samples that contained organisms (Table 23). One lower intertidal sample contained no organisms. The fragments indicated a relatively low standing stock (mean, 33.4 g wet wt/m^2). McRoy (1970) estimated the standing stock of eelgrass in Safety Lagoon, which is about 65 km to the west of Golovnin Lagoon, to be over 400 g dry wt/m^2 in September 1967 (Fig. 2 of McRoy 1970; 357 g/m^2 in September 1968, McRoy 1969). We do not have data on the dry weight of the sea grass in our samples, but it is obvious from the wet weights that the standing stock at our site was much less than at Safety Lagoon. Since our samples were taken intertidally and probably near the upper physiological limits of eelgrass and McRoy's samples were probably taken subtidally where eelgrass beds are likely to be more highly developed, our results are not comparable with his. Nevertheless,

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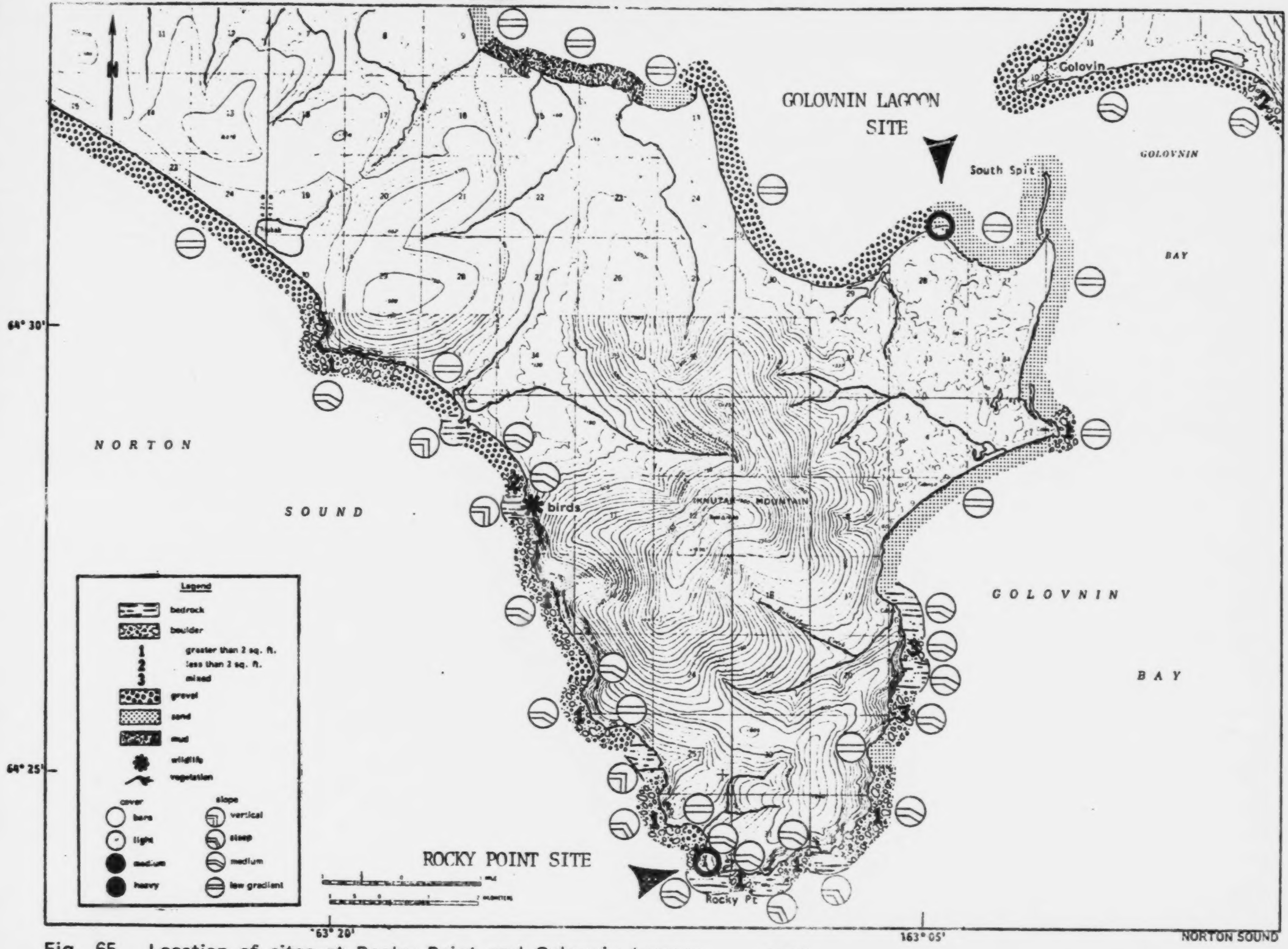
GOLOVNIN LAGOON SITE
Golovin
South Spit
GOLOVNIN
BAY
NORTON
SOUND
birds
IKNUTAK MOUNTAIN
GOLOVNIN
BAY
ROCKY POINT SITE
Rocky Pt
64° 30'
64° 25'
163° 20'
163° 05'
NORTON SOUND
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 65. Location of sites at Rocky Point and Golovnin Lagoon.

Fig. 66. Study site at Golovnin Lagoon. 11 August 1976.

Table 23. Abundances of plants and invertebrates in eight 7-liter samples from a sand beach at Golovnin Lagoon, Norton Sound.

		Abundance[a]							
	Sample no.	H-3	H-2	H-1	M-1	M-2	M-3	L-2	L-3
Species	Elevation (cm)	94	91	76	73	70	67	6	6
Oscillatoriaceae		<0.01	<0.01		0.03	<0.01	<0.01		
Rhodomelaceae						0.5			
Seagrass fragments		0.02	0.2	0.2	2.2	11.0	4.8	0.1	0.2
Oligochaeta		5	8		6	3	7		1
Gammarus sp.					1		3		
Insect larvae							1		

a. Number of individuals of animals, wet weight (g) of plants.

McRoy (1970) concluded from his comparison of eelgrass beds at 10 locations throughout Alaska that standing stock did not appear to change along a latitudinal gradient of increased environmental rigor, but depended instead on "conditions of the local environment." Therefore, one would not necessarily expect two sites at the same latitude to have comparable standing stocks.

The most frequently occurring invertebrates in the sand samples were Oligochaeta (Table 23). The samples from the shore of the bay contained filamentous blue-green algae (Family Oscillatoriaceae), mysids (Neomysis sp.), and midges (Family Chironomidae). Small amounts of sea grass (0.07 g wet weight) were present in one of the three cores.

3.6 Rocky Point

Rocky Point marks the southwestern limit of Golovnin Bay at its entrance (Figs. 55 and 65). We sampled a gradually sloping beach composed of sand at upper levels with increasing amounts of gravel toward lower tidal levels (Fig. 67, Table 24).

A total of six samples was collected at haphazardly selected points in three tidal zones: three samples in the high zone, one in the mid zone, and two in the low zone. For each sample the substrate was shoveled into an 8-liter bucket.

The samples contained few organisms (Table 24). Biota excluded from Table 24 include unidentified colonial diatoms, filamentous algae fragments, and polychaete fragments.

A rocky area adjacent to the sand beach was examined for intertidal biota by S. Zimmerman (unpublished data, 1976, on file Auke Bay Laboratory, P.O. Box 155, Auke Bay, Ak. 99821). Most of it was bare, but small 2-10 mm barnacles (probably Chthamalus dalli) formed a patchily dense cover (100% cover in some patches) in the tidal range from low water to +60 cm. Fucus

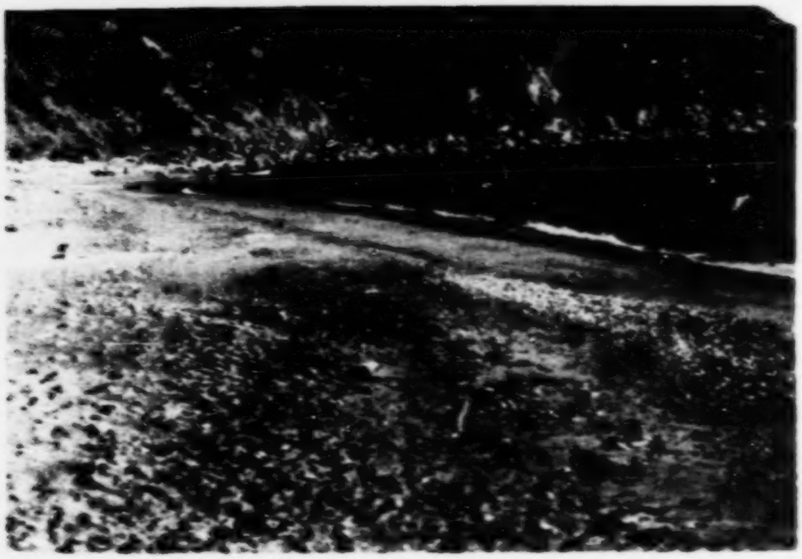

Fig. 67. Study site near Rocky Point. 11 August 1976.

Table 24. Abundances of plants and invertebrates in five 8-liter samples from a sand beach at Rocky Point, Norton Sound.

		Abundance[a]				
	Sample No.	H-2	H-1	H-3	M-1	L-1
Species	Elevation(cm)	119	113	107	52	6
Seagrass fragments			0.07		0.02	
Phyllodocidae					1	
Naineris quadricuspida					1	
Cirratulus cirratus					1	
Tharyx sp.				1		
Oligochaeta					1	
Nematoda						1
Leucon nasica						1
Insecta			1			
Hemiptera		<0.01[b]				

a. Number of individuals of animals, wet wt. (g) of plants.
b. Wet wt. (g). Individuals not counted.

sp. was uncommon; Mytilus was rare. The general pattern of distribution of intertidal organisms was similar to that described below for Sledge Island.

3.7 Square Rock

Square Rock is about 15 km to the east of Topkok Head in northcentral Norton Sound (Figs. 55 and 68). We sampled a gradually sloping gravel beach (Fig. 69). Samples were collected from high-, middle-, and low-tidal zones. In each of the two upper zones, two 8-liter samples each comprising eight haphazardly placed 1-liter cores were collected (Table 15). One semi-quantitative sample about 8 liters in size was collected in the low zone.

Table 25 shows the distribution and relative abundance of eleven invertebrates in three samples from Square Rock. One of the two remaining samples contained no organsisms; the other contained only fragments of filamentous green algae covered with diatoms. The three samples shown in Table 25 also contained fragments of algae, ectoprocts, and Crustacea.

Samples from the highest tidal zone contained oligochaetes, insects, and the partial remains of a barnacle; samples from the middle and low-tidal zones contained errantiate (Eteone longa) and tubicolous Pygospio and Amphiglena polychaetes, insects, and nestling gammarids Anisogammarus and Paramoera.

3.8 Bluff

Bluff is about 10 km east of Topkok Head in northcentral Norton Sound (Figs. 55 and 68). We sampled a moderately sloping mixed gravel and sand beach to the west of the steep bluffs (Fig. 70).

As at most other sites in Norton Sound, we collected composite samples from high-, middle-, and low-tidal zones. In each zone, we collected two 7-liter samples, each composed of seven haphazardly placed 1-liter cores. All were surface cores except two which were deep cores taken in the low tidal zone.

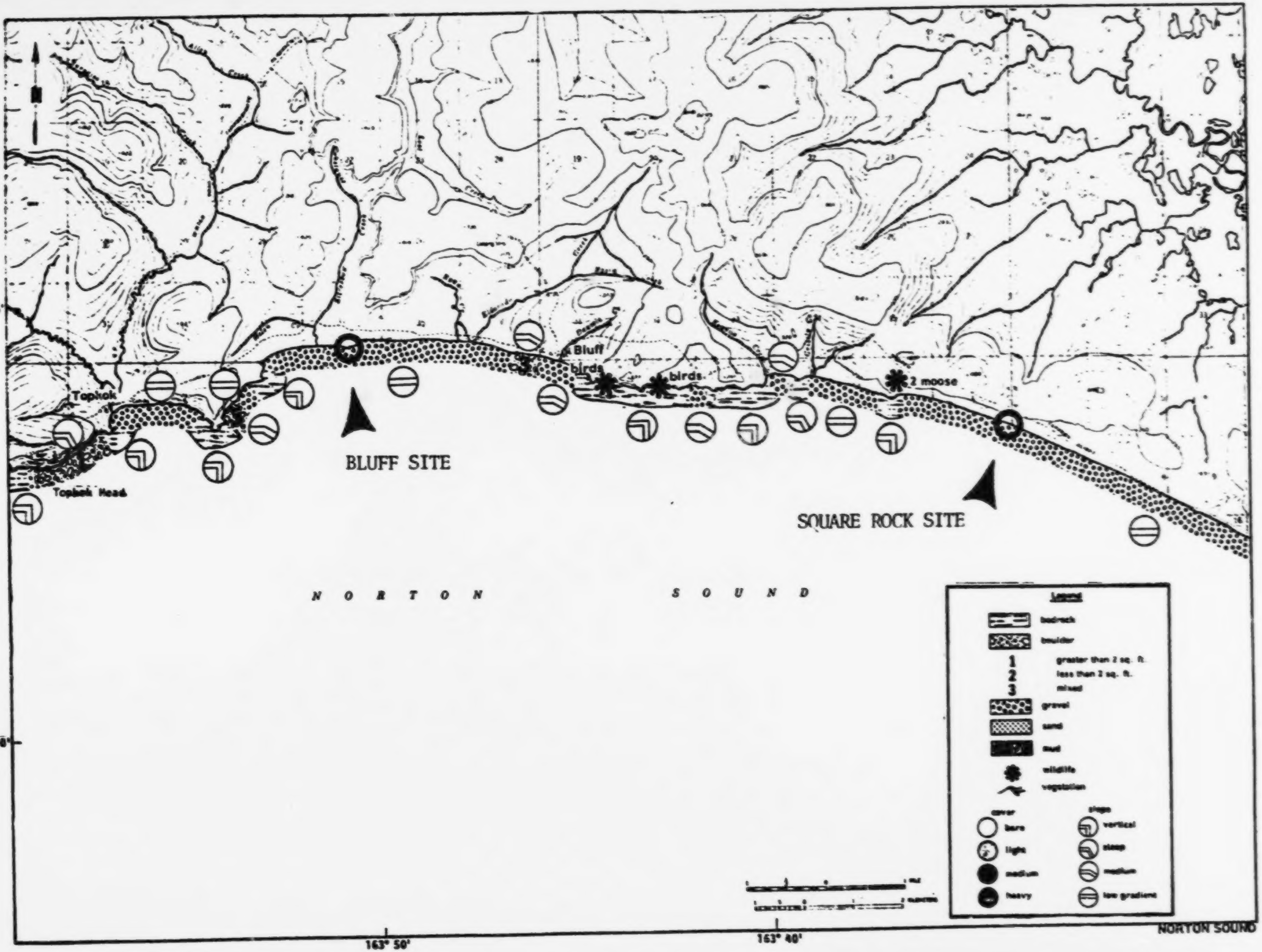
N
4° 35'
14° 30'
Topkok
Topkok Head
Bluff
birds
birds
2 moose
BLUFF SITE
SQUARE ROCK SITE
N O R T O N
S O U N D
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
163° 50'
163° 40'
NORTON SOUND

Fig. 68. Location of sites at Bluff Point and Square Rock.

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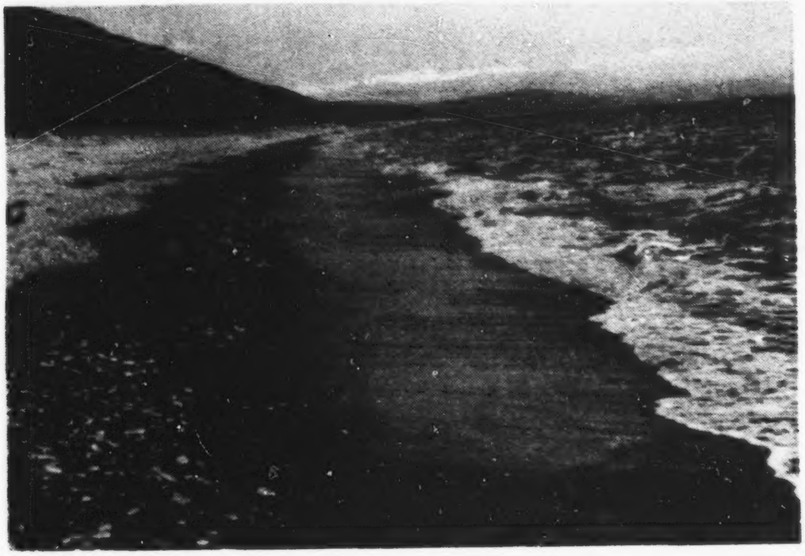


Fig. 69. View to the east from the study area at Square Rock. 12 August 1976.

Table 25. Abundances of invertebrates in three 8-liter samples from a gravel beach at Square Rock, Norton Sound.

		Number of Individuals		
	Sample no.	H-1	M-1	L-1
Species	Elevation (cm)	119	67	15
Eteone longa				1
Pygospio sp.				1
Amphiglena sp.			1	
Oligochaeta		2		
Cirripedia		1[a]		
Amphipoda A			1	
Amphipoda B			1	
Anisogammarus sp.				1
Paramoera columbiana				1
Psyllidae		5		1
Hymenoptera		1		1

a. Shell absent.

Fig. 70. View east from the study site at Bluff. 12 August 1976.

Oligochaetes, hemipterans, and homopterans were the predominant organisms in upper and middle intertidal samples (Table 26). The two Mytilus in sample AH-1 were less than 1.5 cm long and had probably set since the previous winter. Samples from the low tidal zone contained a small amount of brown algae (Order Dictyosiphonales), sessile scyphozoans (Haliclystus sp.), and polychaetes (Eteone longa and Asabellides sibirica). Unidentifiable fragments of red and filamentous green algae occurred in four samples at and below the tidal elevation of +168 cm, but are not shown in Table 26.

3.9 Safety Sound

Safety Sound (Lagoon) is an enclosed body of water along the northwestern shore of Norton Sound (Figs. 55 and 71). Our sampling site was a gently sloping sand and mud beach just east of the entrance to Safety Sound (Fig. 72; Table 15).

Eight semi-quantitative samples, each with a volume of about 8-liters, were collected; one sample represented the high-tidal zone, three were from the middle-tidal zone, and four were from the low-tidal zone. Samples from the low and middle zones were collected under water.

Six samples contained whole organisms (Table 27). The sample collected at the highest tidal level contained only fragments of seagrass, and one sample from the low zone contained no organisms. In addition to the whole organisms shown in Table 27, the samples contained fragments of green algae, polychaetes, and bivalves. Diatoms were epiphytic on the green algae.

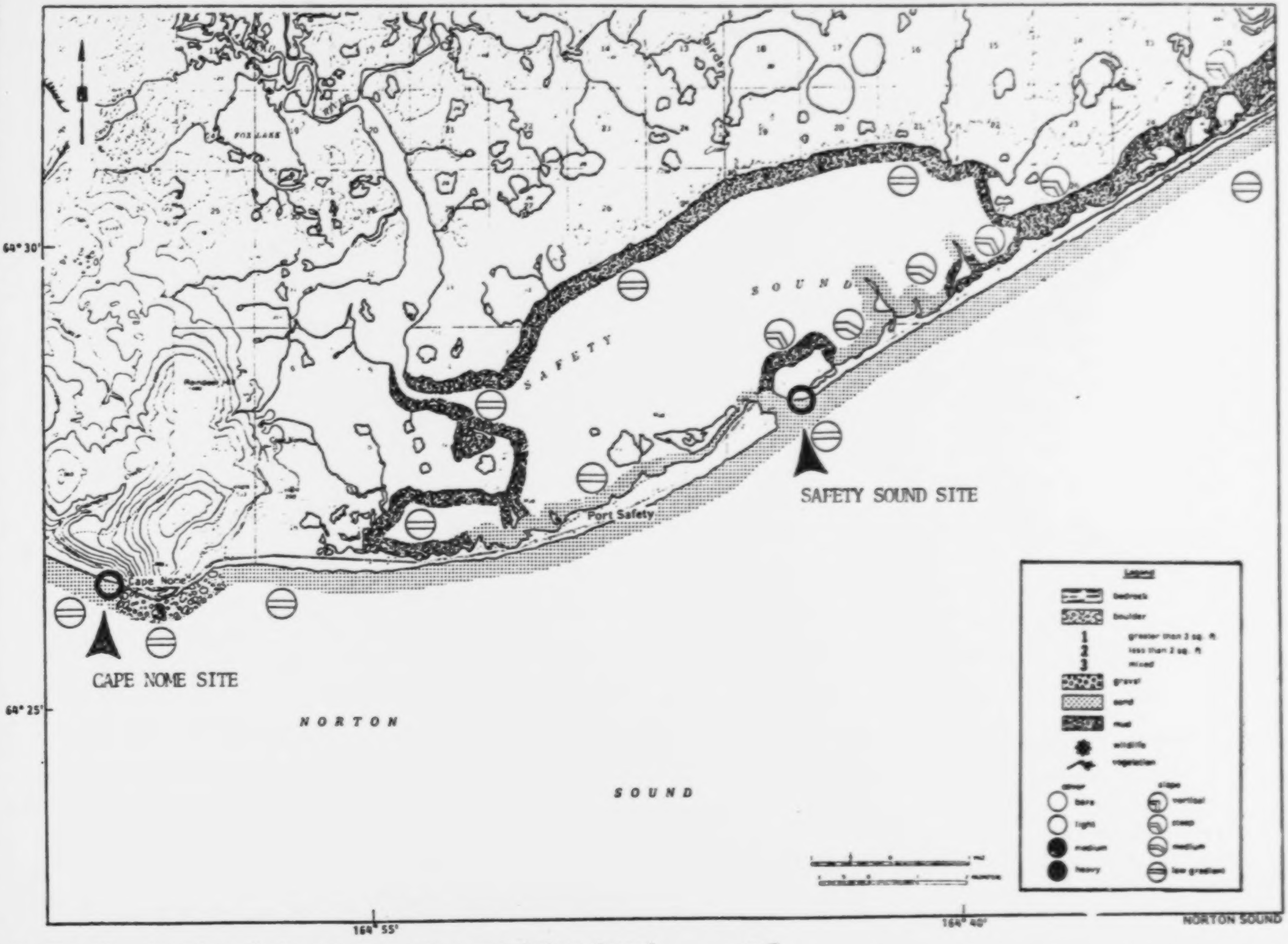
3.10 Cape Nome

Cape Nome is on the northwestern shore of Norton Sound about 20 km southeast of Nome (Figs. 55 and 71). We sampled a wide, gently sloping, sandy beach west of the Cape.

Table 26. Abundances of plants and invertebrates in six 7-liter samples from a gravel and sand beach at Bluff, Norton Sound.

		Abundance[a]					
	Sample no.	BH-1	AH-1	BM-1	AM-1	AL-1	BL-1
Species	Elevation (cm)	186	168	152	116	49	49
Dictyosiphonales							0.7
Gigartinaceae			0.1				
Haliclystus sp.						1	
Polychaeta							1
Eteone longa						1	
Asabellides sibirica							1
Oligochaeta		1	1	3			
Mytilus edulis			2				
Hemiptera				2			
Homoptera		<.01[b]	2		4		

a. Number of individuals of invertebrates, wet wt. (g) of plants.
b. Wet wt. (g). Individuals not counted.



64° 30'
64° 25'
164° 55'
164° 40'
FOX LAKE
SAFETY
SOUND
Port Safety
Cape Nome
SAFETY SOUND SITE
CAPE NOME SITE
NORTON
SOUND
NORTON SOUND
Legend
bedrock
boulder
1 greater than 2 sq. ft
2 less than 2 sq. ft
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 71. Location of sites at Cape Nome and Safety Sound.

269

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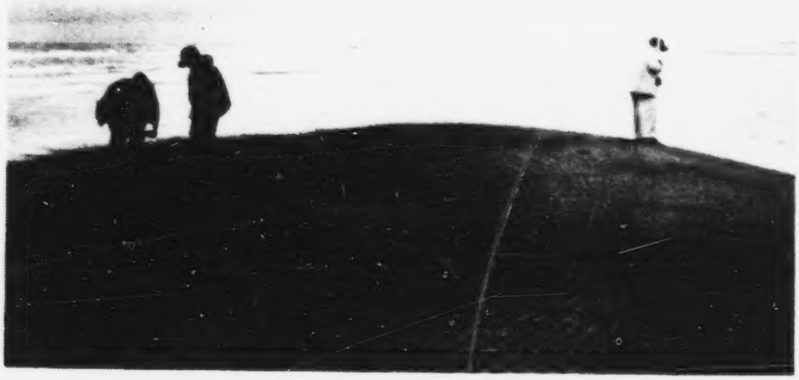

Fig. 72. Study site near the entrance of Safety Sound. 14 August 1976.

Table 27. Abundances of plants and invertebrates in six 8-liter samples from a sand and mud beach at Safety Sound, Norton Sound.

		Abundance[a]					
	Sample no.	AM-1	CM-1	BM-1	AL-1	BL-1	DL-1
Species	Elevation(cm)	-24	-24	-27	-27	-37	-46
Oscillatoriaceae cf. *Lyngbya*							0.3
Bacillariophyceae		<.01		<.01			
Chlorophyta				<.01	<.01		
Rhodomelaceae							.03
Glycinde picta						1	
Pygospio sp.			2	+[b]	3		3
Arenicola glacialis						1	5
Bivalvia					1[c]		
Pontoporeia affinis					1	1	

a. Number of individuals of invertebrates, wet wt. (g) of plants.
b. + = present; no counts made.
c. Shell absent.

The beach was divided into four zones based on the tidal height. From each of two zones, the highest and lowest, we took two 7-liter samples each composed of seven haphazardly placed 1-liter surface. One 7-liter sample collected in the same way was taken from each of the two intermediate zones (Table 15). In addition, two cores (1-liter each) were taken from the lower middle-intertidal zone.

One sample each from the high- and upper-middle zones contained organisms or fragments thereof; the rest of the samples contained no biota (Table 28). Flustrella gigantea had probably washed ashore from sublittoral depths. Feder and Mueller (1974) report Flustrella sp. from trawl samples in the depth range 13 to 23 meters in waters off Nome.

3.11 Sledge Island

Sledge Island is a small island off the southwestern coast of the Seward Peninsula (Figs. 55 and 73). The shore of the island was covered with boulders, except where short stretches of sand and gravel beach intervened. We collected six samples of coarse sand from a beach on the southwest side of the island (Fig. 74). A pair of samples was collected in each of the high-, middle-, and low-intertidal zones between the 18 and 158 cm tide levels. Samples were shoveled into separate 3.5-liter buckets (Table 15).

Few species were found in the quantitative samples, and among these species, worms were most prevalent (Table 29). Oligochaetes occurred in four samples and were most abundant in the mid-intertidal zone. Two species of polychaete and the collembolan Anurida maritima occurred in the mid-intertidal zone samples. The only identifiable alga was a species of Rivulariaceae. Fragments of red algae, ectoprocts, and crustaceans were also found in our samples, probably cast up from the subtidal region.

Table 28. Abundances of plants and invertebrates in two 7-liter samples from a sand beach near Cape Nome, Norton Sound.

		Abundance[a]	
	Sample no.	BH	U
Species	Elevation (cm)	28	6
Oscillatoriaceae cf *Lyngbya*		0.1	
Sphacelaria racemosa		0.05	
Rhodomelaceae		0.06	
Potamogetonaceae		3.1[b]	0.05[b]
Flustrella gigantia		0.2	
Onisimus litoralis			1

[a] Number of individuals of *Onisimus*, wet wt. (g) of plants and *Flustrella*.

[b] Fragments.

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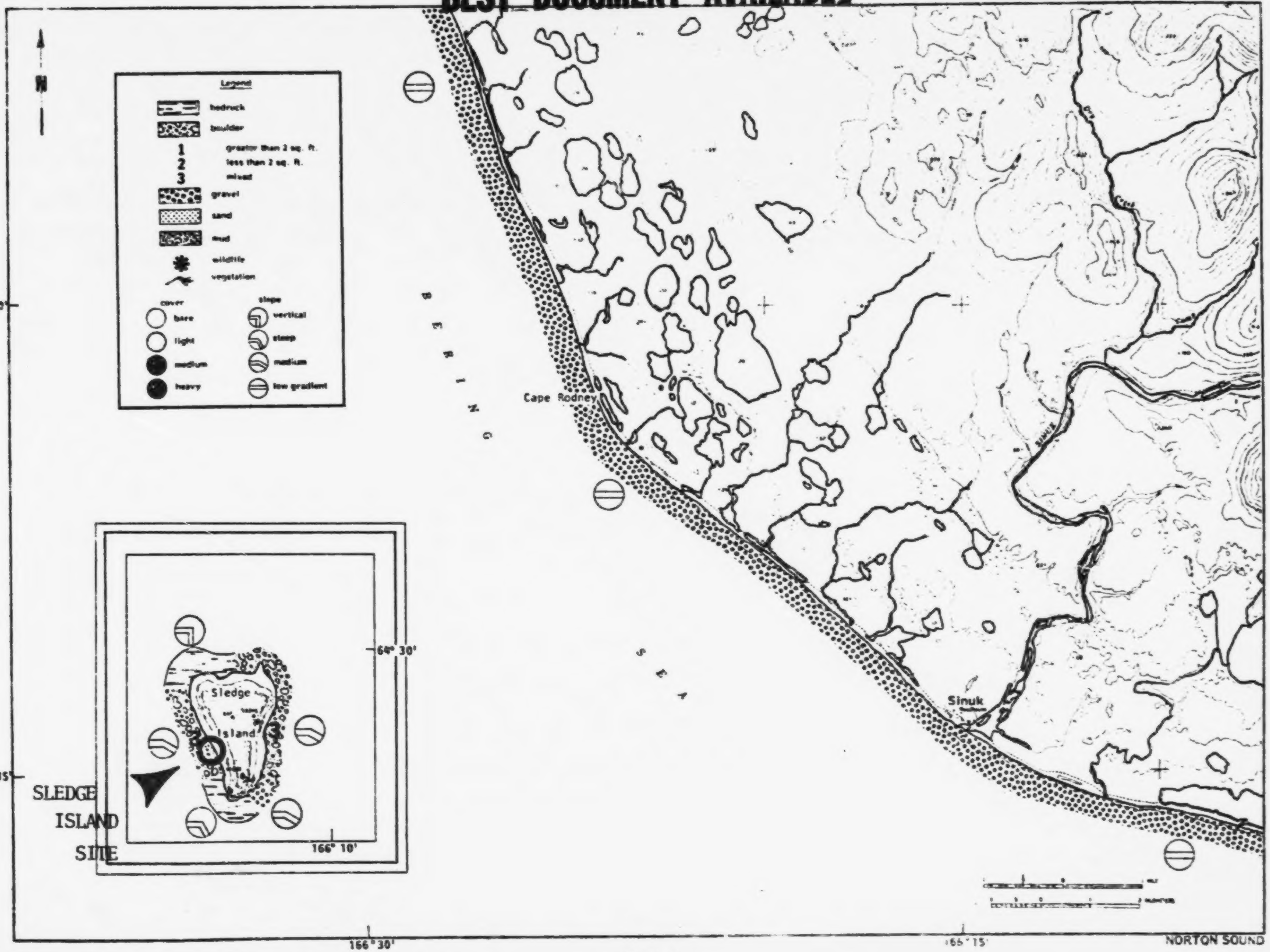

N
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient
64°40'
64°35'
B E R I N G
S E A
Cape Rodney
Sinuk
Sledge Island
64° 30'
166° 10'
SLEDGE ISLAND SITE
166°30'
165°15'
NORTON SOUND

Fig. 73. Location of the site at Sledge Island.

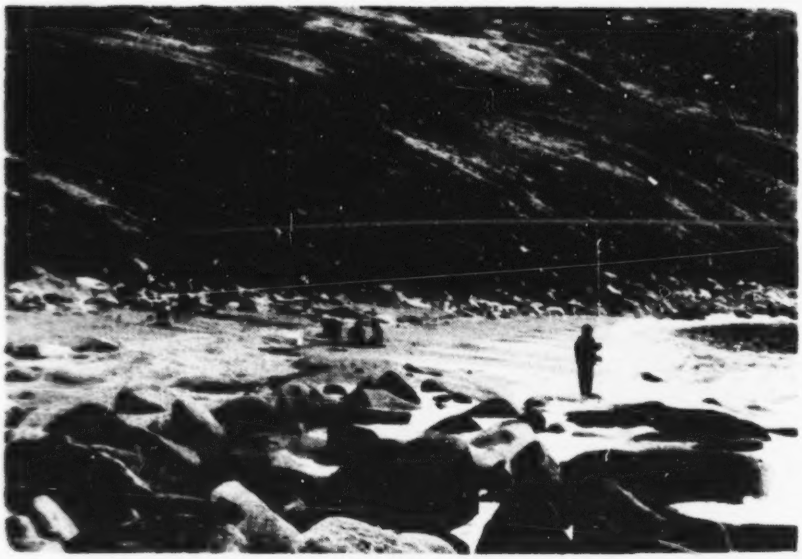

Fig. 74. Sledge Island study site, Norton Sound. 7 August 1976.

Table 29. Abundances of species of algae and invertebrates in samples from a sand beach at Sledge Island, Norton Sound.

				Abundance[a]			
	Sample No.	H-1(A)	H-1B	M-1A	M-1B	L-1A	L-1B
Species	Elevation (cm)	158	158	98	94	18	18
Rivulariaceae cf. *Rivularia*					.005		
Eteone longa						2	
Saccocirrus sp.						47	32
Oligochaeta			3	88	49		2
Anurida maritima					18		
Diptera				2L[b]	2		
Culicidae		1					

a. Wet weight (g) of plants, number of individuals of animals.
b. L = larvae.

We also made qualitative observations of the biota at a boulder area near the sampling site. The tops of the boulders were nearly bare, but we noted clumps of Fucus in crevices (Fig. 75) and patches of small (5 to 10 mm) barnacles. All sessile organisms occurred below MLLW, and only littorine snails were seen above this level (Fig. 76).

3.12 Cape Woolley

Cape Woolley is on the southwestern shore of the Seward Peninsula about 55 km northwest of Nome (Figs. 55 and 77). We sampled a moderately sloping sand beach to the north of the bedrock headland at Cape Woolley (Fig. 78) and made observations of the biota on rock at the headland itself.

The sand beach was divided into high, middle, and low zones. From each zone, we collected two samples, each composed of seven haphazardly placed 1-liter surface cores (Table 15). In addition, two 7-liter samples composed from deep cores were taken in the middle and high zones.

Seven samples contained whole organisms or fragments thereof. Of these, three contained only fragments of algae, seagrass, hydroids, ectoprocts, isopods, and insects. Two samples taken from 10 to 20 cm below the surface of the sand contained only the remains of hydroids, ectoprocts, and insects. Ectoproct fragments occurred more frequently than other biotic remains, being present in six of the seven samples. As with hydroid fragments, the ectoprocts had probably washed ashore from subtidal depths. Barr (in Zimmerman et al. 1977) observed both groups at a depth of 55 ft (16.8 m) four miles (6.4 km) offshore from Cape Woolley. The abundances of organisms in those samples containing whole organisms are shown in Table 30.

At the headland, Zimmerman saw small patches of Fucus on inaccessible offshore rocks, and south of the headland he noted complete coverage of a rocky area by Chthamalus, Fucus, and diatoms (Fig. 79). He also indicated

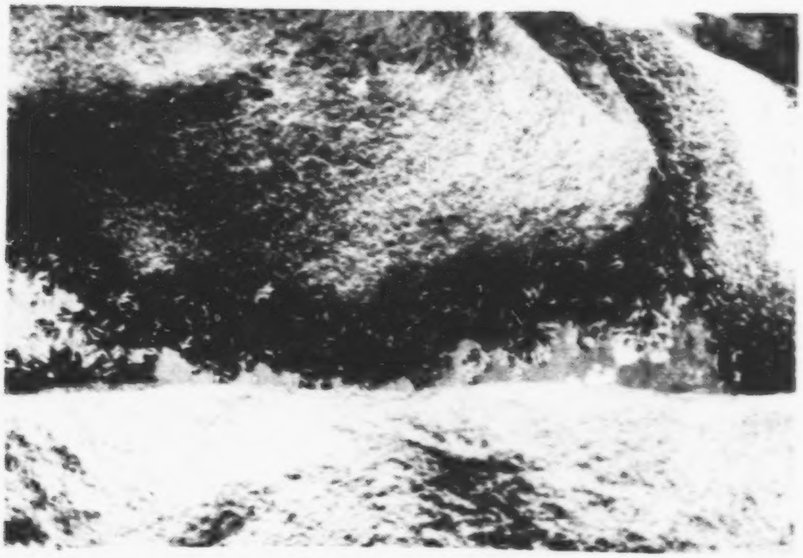

Fig. 75. Fucus sp. growing between boulders below MLLW. All sessile organisms were restricted to such sheltered habitats on the boulder beach at Sledge Island.

Fig. 76. Littorina sp. grazing a patch of algae (dark area in center) on boulder at Sledge Island.

The shop seemed to be full of all manner of curious things--but the oddest part of it all was that, whenever she looked hard at any shelf, to make out exactly what it had on it, that particular shelf was always quite empty, though the others round it were crowded as full as they could hold.

"Things flow about so here!" she said at last in a plaintive tone, after she had spent a minute or so in vainly pursuing a large bright thing, that looked sometimes like a doll and sometimes like a work-box, and was always in the shelf next above the one she was looking at. "And this one is the most provoking of all--but I'll tell you what--" she added, as a sudden thought struck her. "I'll follow it up to the very top shelf of all. It'll puzzle it to go through the ceiling, I expect!"

But even this plan failed: the "thing" went through the ceiling as quietly as possible, as if it were quite used to it.

Through the Looking Glass
Lewis Carroll

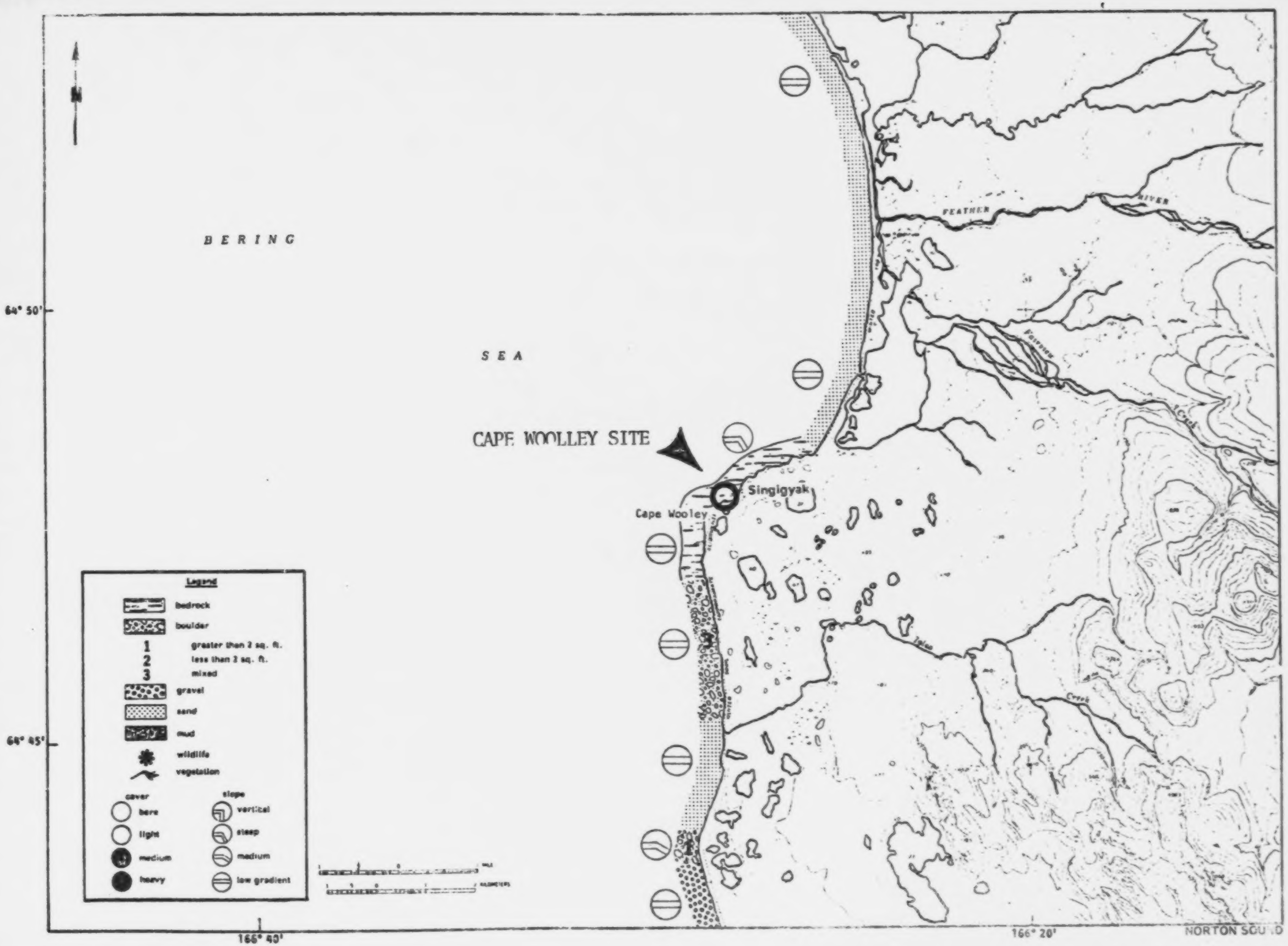
N
BERING
SEA
CAPE WOOLLEY SITE
Singigyak
Cape Wooley
FEATHER
RIVER
Creek
64° 50'
64° 45'
166° 40'
166° 20'
NORTON SOUND
Legend
bedrock
boulder
1 greater than 2 sq. ft.
2 less than 2 sq. ft.
3 mixed
gravel
sand
mud
wildlife
vegetation
cover
bare
light
medium
heavy
slope
vertical
steep
medium
low gradient

Fig. 77. Location of site at Cape Woolley.

Fig. 78. Study site on a sand beach north of Cape Woolley. 13 August 1976.

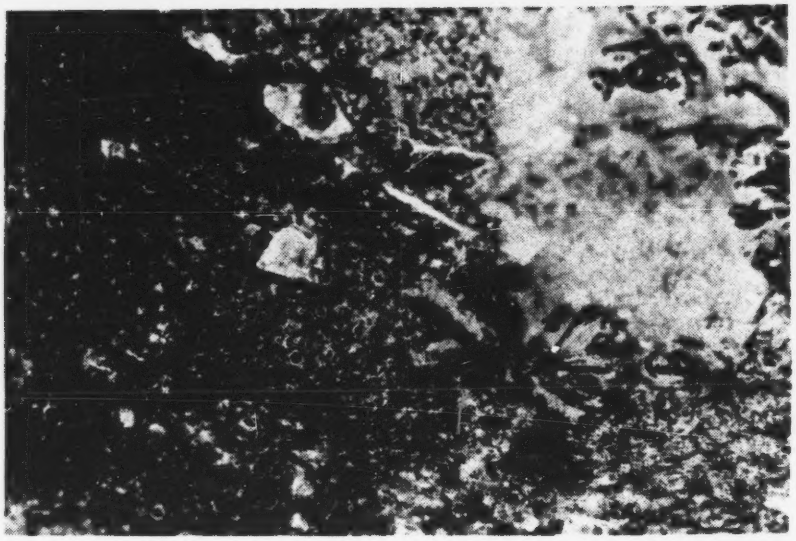

Fig. 79. Rock at Cape Woolley covered with Fucus, Chthamalus, and an unidentified filamentous alga. 15 August 1976.

Table 30. Abundances of plants and invertebrates in four 7-liter samples from a sand beach at Cape Woolley.

		Abundance[a]			
	Sample no.	AH-1	BH-1	BM-1	AL-1
Species	Elevation(cm)	131	94	91	34
Cyanophyta		<.01f[b]		<.01	
Fucus sp.		<.01f			
Capitella capitata			1		
Cirratulus cirratus			1		
Gammarus sp.					1
Hymenoptera		.01[c]			
Culicidae					1

a. Number of individuals of animals, wet wt. (g) of plants.
b. f = fragment.
c. Wet wt. (g). Individuals not counted.

that intertidal tidepools contained only diatoms and that the drift biota included zooecia and shells of ectoprocts and Mytilus, respectively.

3.13 King Island

King Island is a small rocky island in the northern Bering Sea 53 km west of Cape Douglas on the Seward Peninsula (Fig. 80). Cliffs rise precipitously from the sea here and provide only a narrow intertidal zone occupied by very large boulders (Fig. 81). We made no intertidal collections because few macroscopic organisms were present (Fig. 82). The boulders at the water's edge were covered with a slippery film of green or blue-green algae; small patches of Halosaccion glandiforme occurred in depressions and cracks in the rock surface. Recent scouring of the intertidal area by ice was presumably responsible for the extremely low species diversity of intertidal organisms.

Adjacent to our intertidal site, divers made observations of the sublittoral region to a depth of 25 m (Zimmerman et al. 1977). They recorded a diverse benthic biota including starfish (Crossaster and Leptasterias), basket stars (probably Gorgonocephalus), crabs (Hyas and Paralithodes platypus), tunicates (Boltenia), and abundant kelps (Laminaria, Alaria, and Agarum). The neritic region was also richly populated with invertebrates. The divers observed evidence of disturbance of the substrate by ice down to a depth of at least 26 m.

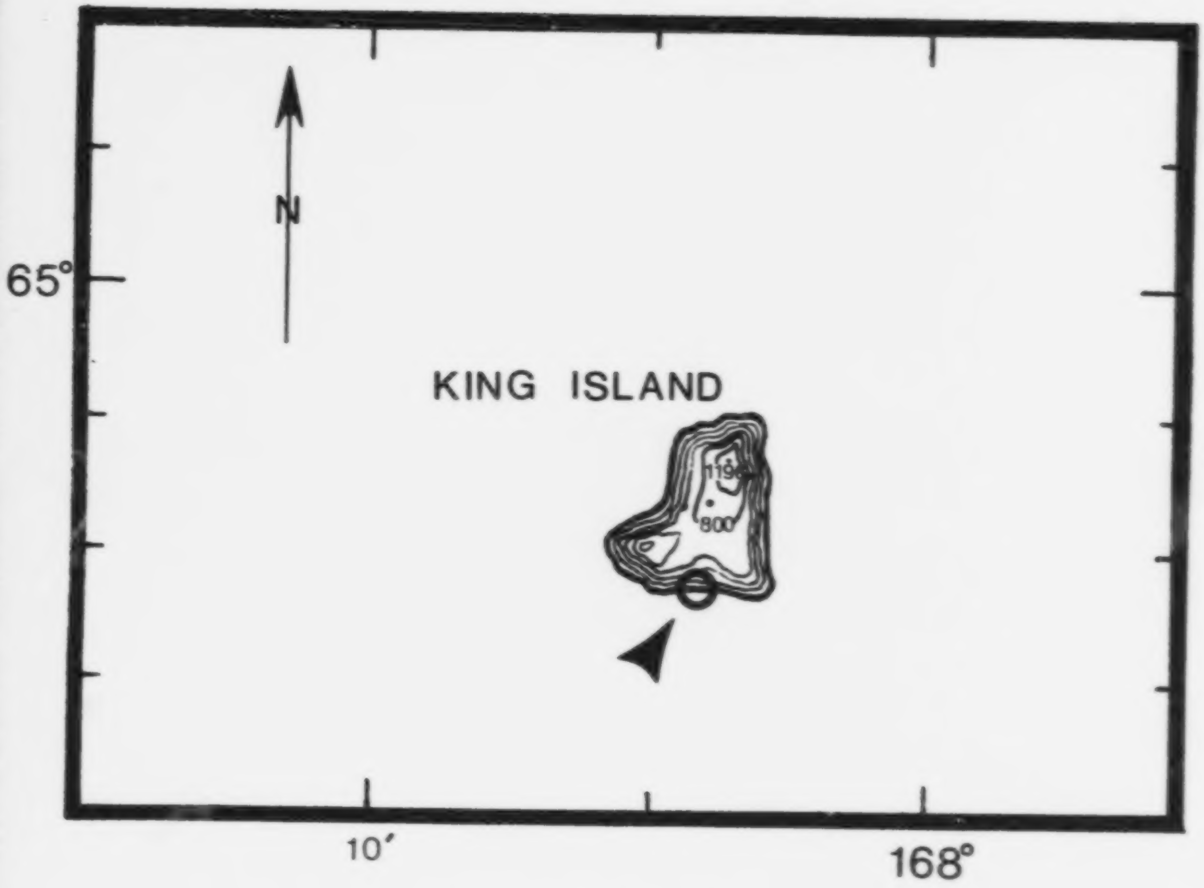
N
65°
KING ISLAND
119
800
10′
168°

Fig. 80. Location of the study site at King Island.

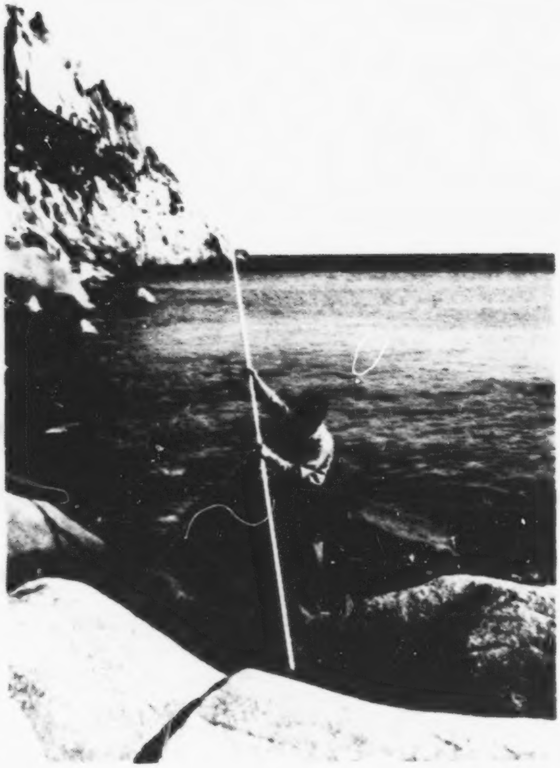

Fig. 81. Boulder-strewn intertidal area at King Island. Biological cover consisted almost exclusively of a thin film of green or blue-green algae. 6 August 1976.

Fig. 82. Intertidal sampling area below abandoned village at King Island. Random sampling was begun but was discontinued because of the paucity of intertidal biota. 6 August 1976.

B. Effects of Scouring by Sea Ice on Intertidal Community Structure

1. Physical Disturbance by Sea Ice

The Pribilof Islands are near the southern limit of sea ice in the Bering Sea. Wise and Searby (1977) summarize observations on the distribution and coverage of sea ice in the Bering Sea for the period 1954 through 1970. They show the Pribilof Islands at the southernmost latitude of 15-day means of the edge of the pack ice from February through April. Amak and Akun Islands are shown south of the extreme southern limit of the pack-ice edge in all months of the year, although Amak Island is shown near this limit from February through April.

We used information on the distribution, coverage, and thickness of sea ice in the Bering Sea that was contained in weekly summary ("Southern Ice Limit") or "30 Day Sea Ice Forecast" charts of the Navy Fleet Weather Facility to determine sea ice conditions in the southern Bering Sea in five winters (1972-76) immediately preceding and during our field studies there (Table 31). The charts were drawn primarily from satellite imagery supplemented by conventional observations.[1] The records that were available to us at the time of this writing for the years prior to 1972 were inadequate for determining ice conditions in the vicinity of the Pribilof and Amak Islands.

The ice charts show that the Pribilof Islands were surrounded by pack ice in four out of five winters immediately preceding and during our field studies (Table 31). In the winter of 1973 when the Pribilofs were ice free, St. Paul Island was very near the southern limit of ice in late April. The southern limit of ice is shown in the vicinity of Amak in 2 years (1974 and 1976), but the small scale of the charts did not allow a determination of whether the ice reached Amak.

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[1] Conventional observations include those obtained from ships, shore stations, and aerial reconnaissance.

Table 31. Sea Ice at the Pribilof Islands and Amak Island in Five Recent Winters.[a]

	Pribilof Islands			Amak Island		
Year	Dates of first and last ice[b]	Total days in ice	Highest coverage (Oktas)[c]	Dates of first and last ice[b]	Total days in ice	Highest coverage (Oktas)
1976	24 Jan., 4 May	20(8)[d]	not available	27 Mar., 27 Apr.	8	7-8
1975	18 Jan., 30 Mar.	23(14)[c]	6-8	ICE FREE		
1974	26 Feb., 30 Apr.	7(31)	4-6	26 Feb., 12 Mar. (Amak at ice edge)	15	5-7
1973	ICE FREE (St. Paul near ice edge on 24 April 1973)			ICE FREE		
1972[e]	20 Mar., 5 Apr.	(17)	7-8	ICE FREE		

a. Data from weekly summary, "Southern Ice Limit", or "30 Day Sea Ice Forecast" charts of the Fleet Weather Facility, U.S. Navy, Suitland, Maryland.
b. Islands were not necessarily iced in during entire period between first and last dates.
c. Amount of ice cover in eighths.
d. Number in parentheses indicates added days St. Paul and Otter Islands but not St. George Island were in ice.
e. Data for 1972 available to us at the time of this writing were incomplete.

The most important effect of pack ice on intertidal communities of organisms is probably scouring of the substrate. In the Pribilof Islands ice scouring is a frequent and probably widespread and severe physical disturbance to intertidal communities. Pack ice appears to be nearly an annual occurrence in the Pribilofs, at least in recent years, and is therefore frequent with respect to the life span of most of the ecologically important inhabitants.

Sea ice occurs at the Pribilof Islands in late winter and early spring (Table 31). Although extreme spring tides occur there in early winter and early summer, tidal fluctuations during the period of sea-ice coverage result in most intertidal levels being subject to ice scouring. Moreover, when the islands are surrounded by ice, there are presumably few stretches of shoreline unaffected by scouring. Scouring, therefore, is a widespred disturbance both horizontally and vertically in the Pribilofs.

Many of the dominant organisms in intertidal communities are sedentary and therefore cannot retreat into crevices in bedrock or the interstices of boulder fields or migrate to lower levels to avoid being crushed or scraped from the substrate by sea ice. The removal of large numbers of dominant organisms from surfaces exposed to scouring would severely disturb the organization of an intertidal community.

In theory, disturbances that are frequent, widespread and severe should simplify communities by reducing species richness (Grime 1973, Levin and Paine 1974, Huston 1979). From the above statements, we would expect lower species richness in intertidal communities at the Pribilofs than at nearby islands which are less frequently affected by sea ice.

1.1 Island Biogeography

In terrestrial systems species richness on oceanic islands depends on and is usually reliably predicted by the size of the island and its distance from the nearest mainland area, factors which affect the rates of immigration and extinction of potential island colonists (MacArthur and Wilson 1963, 1967). Although field experiments in the marine environment generally tend to support the MacArthur-Wilson theory of island biogeography, at least one study contradicts the simple linear model (Schoener et al. 1978). Furthermore, all tests of the model were conducted on patches of environment (not true islands) such as plastic mesh sponges (Schoener 1974a), slate, wood or asbestos panels (Schoener 1974b, Osman 1978), and rocks (Osman 1978) none of which exceeded a surface area of 2500 cm^2.

For the purposes of the present discussion, we assume that the effect of island size and distance from the source area (mainland Alaska) on differences in within-habitat species richness among Akun Island, Amak Island, and the Pribilof Islands is negligible compared with the effect of scouring by ice for the following reasons:

1. Dispersal over stretches of ocean is less hazardous for marine propagules (spores, larvae, rafted adults etc.) than for terrestrial ones, therefore, immigration rates of intertidal species to all of the islands should be high and differences in immigration rates to islands at different distances from the source area should be reduced.
2. Oceanic currents may influence the direction and rate of transport of marine propagules so as to reduce differences in immigration rates between source areas and near versus far islands. Surface currents are more likely to influence the dispersal of propagules

from intertidal areas than are subsurface currents. Surface currents in the Bering Sea (Favorite et al. 1976) probably bring marine propagules to the Pribilof Islands primarily from two geographical areas by the way of two routes: the Alaska Peninsula and the eastern Aleutian Islands via the Transverse Current of the Bering Current System, and the shores of Bristol and Kuskokwim Bays via the Pribilof Current (Favorite et al. 1976). Both routes provide reasonably direct transport of propagules to the Pribilof Islands from the nearest source areas.

The Bering Current System in the vicinity of Akun and Amak Islands flows eastward, therefore, the most likely source areas of propagules for these two islands are the eastern Aleutian Islands. Another possible source area would be the shores of the western Gulf of Alaska. Propagules from these shores could be transported via branches of the Alaskan Stream through the Aleutian Passes and eventually to Akun and Amak Islands. Whether propagules arrive from the Aleutian Islands or from the western Gulf of Alaska, we would expect a lower equilibrium number of species on Akun and Amak Islands than if dispersal were directly west to these islands from the Alaska peninsula because, in the first case, the species pool on the Aleutian Islands is likely to be smaller than that on the Peninsula and, in the second case, the effective distance between source area and the islands is greater, and, consequently, immigration rates are lower. As a result, we would not expect the equilibrium number of species on Akun and Amak Islands to differ as greatly from that of the Pribilof Islands as it would if currents were ignored and only straight line distances to nearest major source area were considered.

3. The amount of rocky intertidal habitat (area of shore) on even the smallest island that we consider is very large compared with the body size of most intertidal organisms. Extinction rates should be low and probably similar for all of the above islands. This argument may not hold if the probability of extinction of a keystone species (which tend to be larger than most other animals in a community) was significantly greater on smaller islands since these species have a profound effect on community composition at the local level (as has been shown experimentally by Paine [1966, 1971] and Addicott [1974] and has been suggested for certain marine systems by Mann and Breen [1972], Estes and Palmisano [1974] and Palmisano and Estes [1977]).

The latter two authors studied the effect of predation by sea otters on populations of sea urchins and the resulting effect on the structure of nearshore communities in the Aleutian Islands. Although they did not examine the question of whether biogeography or predation was more important in determining the composition of the nearshore community on the islands they studied, their results indicate that the effect of predation was probably overriding. In a later section, we note the relatively high species richness in refuges from ice scouring at the Pribilof Islands. This evidence supports our assumption that ice scouring outweighs biogeography in determining community composition at the Pribilof Islands.

In order to examine the effect of ice scouring on intertidal community structure, we used two community attributes, 1) species richness as approximated by average species densities (number of species in 1/16 m^2 quadrats) and species-area (sample size) curves, and 2) species-importance curves, to compare intertidal communities at two sites in the Pribilof Islands

(Otter Island and Zapadni Bay, St. George Island; Fig. 1) with those at a site each at Amak and Akun Islands. Data collected in July and August 1975 were used in the analyses. The intertidal region at each site was divided into upper (between mean high water [MHW] and mean tide level [MTL]) and lower (between MTL and mean low water [MLW]) intertidal zones after the scheme proposed by Rigg and Miller (1949).

Species in most major taxa of plants and invertebrates are considered. The following taxa are excluded because organisms from them were usually not identified below the level of order: Porifera, Cnidaria, Platyhelminthes, Nemertea (except *Emplectonema gracilis*), Oligochaeta, Copepoda, Tanaidacea, Insecta (except *Anurida maritima*), Arachnida, Acarina, Sipuncula, Bryozoa and Ascidiacea.

1.2 Species Richness

Average numbers of species in the Pribilof Island samples ranged from one half to less than a quarter of those at Amak and Akun Islands (Table 32). We tested the significance of these results with a 2-way analysis of variance (anova). Since the cell means were approximately equal to their respective variances, we transformed ($[x + 0.5]^{\frac{1}{2}}$) the counts. Bartlett's test revealed that the variances of the transformed counts were homogeneous ($p = 0.62$).

The anova revealed that the treatment means for sites and levels were not from the same population (Table 33). The interaction term was not significant, so orthogonal comparisons were made of means of counts in the upper and lower intertidal zones combined (Table 34). The mean of counts from the Pribilofs (St. George and Otter Islands combined) was significantly less than ($p < 0.001$) that for Amak and Akun Islands combined. There was no significant difference in counts between St. George and Otter Islands, but

Table 32. Average species counts of plants and invertebrates in quadrats ($1/16\ m^2$) at rocky intertidal areas in the Pribilof Islands and at Amak and Akun Islands.

Site	Upper intertidal area			Lower intertidal area		
	n	$\bar{x}$	S.D.	n	$\bar{x}$	S.D.
St. George Island (G)	2	0.5	0.7	4	7.2	3.4
Otter Island (O)	3	3.7	1.5	18	6.7	2.6
Amak Island (Am)	2	13.5	7.8	5	13	3.8
Akun Island (Ak)	3	21	2.6	10	32.4	9.4

Table 33. Analysis of variance of species counts of algae and invertebrates at upper and lower rocky intertidal levels in the Pribilof Islands and at Amak and Akun Islands. ** = highly significant; n.s. = not significant.

Source	d.f.	SS	MS	F
Intertidal level	1	5.20	5.20	15.33**
Site	3	53.42	17.81	52.55**
Level X Site	3	0.85	0.28	0.84 n.s.
Error	39	13.21	0.34	

d.f. = degrees of freedom.

SS = sum of squares.

MS = mean squares.

F = f-distribution value.

Table 34. Three orthogonal comparisons of species counts of algae and invertebrates in the rocky intertidal region at the Pribilof Islands and at Amak and Akun Islands. ** = significant; n.s. = not significant.

Comparison[a]	Difference of means	SS	F
G vs. O	0.41	0.79	2.32 n.s.
G O vs. Am Ak	4.39	44.41	131.08**
Am vs. Ak	1.80	14.67	43.28**

a. G = St. George Island, O = Otter Island,
Am = Amak Island, Ak = Akun Island.

SS = sums of squares.

F = f-distribution value.

counts at Amak were significantly less than those at Akun ($p < 0.001$; Table 34).

Amak Island is shown south of the southernmost limit of pack ice on the charts of Wise and Searby (1977), but data from the Navy Fleet Weather Facility indicate that sea ice may have affected the intertidal area there in late winter 1974 (Table 31). Nevertheless, because of the infrequent occurrence of sea ice at and the disparity in sizes of Amak and Akun Islands, reduced species richness in the intertidal region at Amak may reflect a lower equilibrium number of species (assuming that communities there are at equilibrium) resulting from a higher rate of extinction of colonists.

1.3 Species-area Curves

We chose Mollusca to compare species-area and species-importance relations (see below) in the intertidal region at Amak, Akun, and the Pribilof Islands because it is a diverse group of invertebrates with representatives at all intertidal levels, and the Bering Sea fauna is comparatively well known taxonomically. Other diverse groups of intertidal organisms such as Rhodophyta, Polychaeta, and Gammaridea are more susceptible to the physiological stresses of the upper intertidal environment, and therefore have few representatives at upper levels. The Bering Sea representatives of these groups are poorly known taxonomically.

Eight quadrats were randomly selected from sampled quadrats which fell in the range MLW to just above MHW at each site. The sample size for each site was limited by the total number of samples (eight) collected in this range at Amak Island. Sample size appeared to be adequate for all sites except Akun Island (Fig. 83). The cumulative species count for Akun Island continued to climb as sample size was increased to eight, but when more samples (seven) were randomly added the species counts eventually leveled

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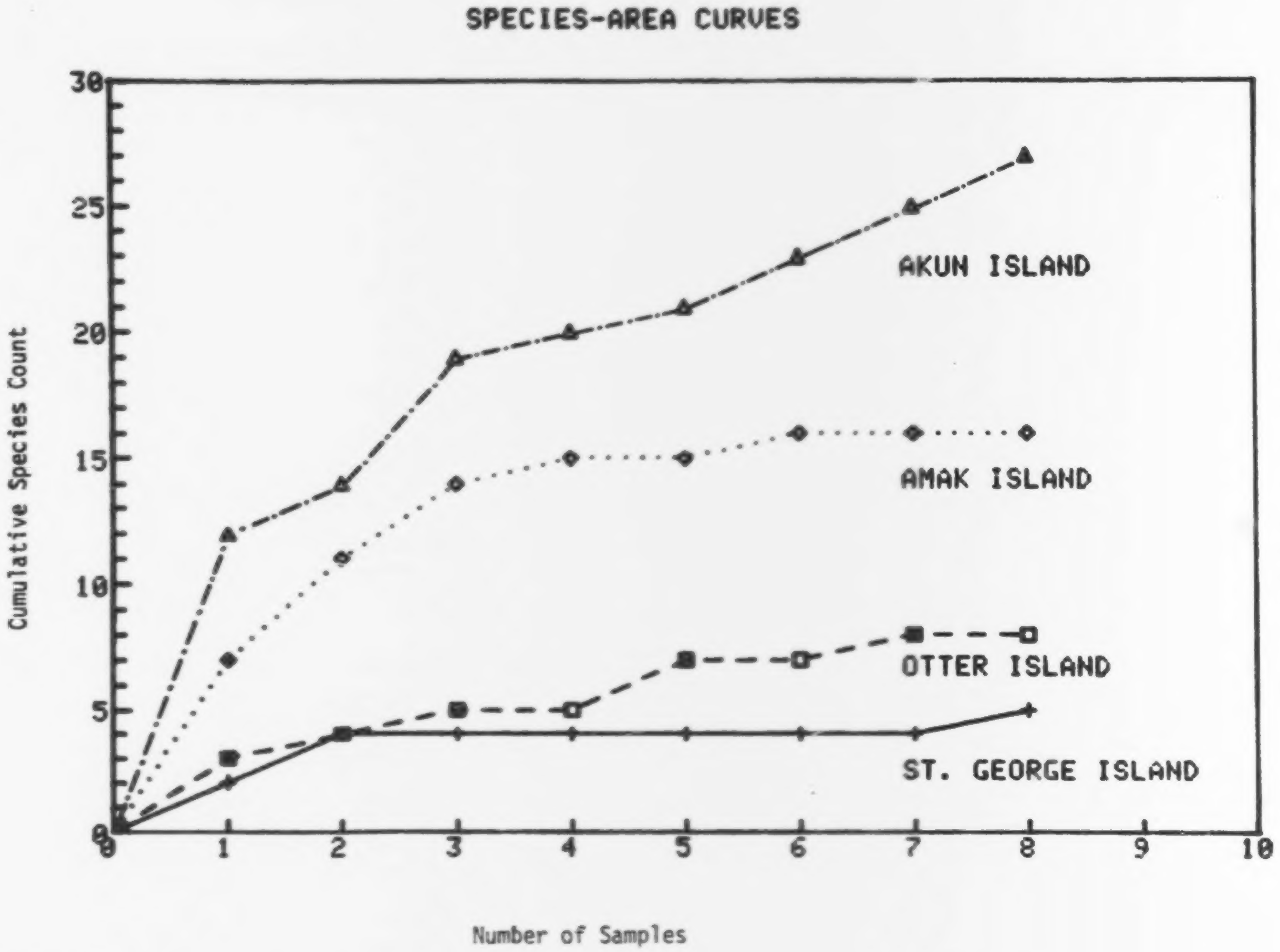

Fig. 83. Species-area curves of mollusks in quadrats collected in the intertidal region at four islands in the Bering Sea. The shores of two islands (Otter and St. George) are frequently scoured by sea ice.

off at 31 species. Therefore, it appears that even at the most diverse island the sample size included 87% of the species of mollusks in the intertidal zone.

The results of the between-island comparisons of mollusk species-area curves paralleled those of the species counts of most major taxa (Fig. 83). The curves for the Pribilof Islands leveled off at the lowest species counts (four to seven) and that for Akun Island increased through the highest count (27) probably leveling off at over 30 species. The curve for Amak reflected an intermediate species richness.

A Smirnov test of the empirical distribution functions of the species area curves revealed no significant difference between St. George and Otter Islands (Table 35). The species-area curve for Otter Island was significantly different in form ($p < 0.001$) than that for Amak Island. Amak Island, in turn, was significantly lower ($p < 0.05$) in mollusk species richness than Akun Island (Table 35).

1.4 Species-importance Curves

We used dominance-diversity curves (Whittaker 1965, 1970, 1972) to examine the distribution of importance among species. These curves are constructed by plotting the importance of a species on the ordinate opposite its respective rank, in terms of the measure of importance selected, on the abscissa. Species are ranked from most to least important on the abscissa. Various authors have used different measures of species importance such as abundance, biomass, coverage, and productivity. Whittaker (1965) has found that in terrestrial plant communities at least three of these measures (coverage, biomass, and production) produce dominance-diversity curves which differ in steepness but not form. Batzli (1969) has claimed that in the rocky intertidal region biomass is a better measure of dominance than the

Table 35. Smirnov test[a] of differences in species-area curves for mollusks in the intertidal region at St. George, Otter, Amak, and Akun Islands. n.s. = not significant.

Contrast	Test statistic[b] T2	Significance
St. George Is. vs. Otter Is.	5/8	n.s.
Otter Is. vs. Amak Is.	7/8	$p < 0.001$
Akun Is. vs. Amak Is.	6/8	$p < 0.05$

a. one-sided K - sample Smirnov test.

b. K = 4, N = 8.

number of individuals. We use biomass (wet weight) as a measure of importance here.

Three theoretical distributions of importance among species occur frequently in the literature; May (1975) relates them. The most uniform is the broken stick distribution and the least uniform is the geometric series. The log-normal distribution falls between these two. Two of the distributions --the broken stick and the geometric series--reflect, in theory, biological mechanisms (types of competition) which are structuring the community. The log-normal distribution arises when species-importance relations are controlled by the "interplay of many independent factors" (May 1975).

Pielou (1975) argues that when the total number of species present is being estimated by the data and the community is small, as is the case in the present study, one cannot test the fit of the observed to the theoretical distribution statistically. However, we can test the difference between the empirical distributions with tests of the Smirnov type (Pielou 1975, Conover 1971).

Visual comparison of the species-importance curves for Amak, Akun, and the Pribilof Islands shows that biomass was most evenly distributed among the species at Akun Island (Fig. 84). The distributions become less and less uniform (i.e. toward a greater concentration of biomass among a few dominant species) as one considers, in turn, Amak, Otter, and St. George Islands. (Fig. 84).

We tested only the curve for St. George Island against that for Akun Island with the Smirnov test because tests of this type which can detect all types of differences that may exist between empirical distribution functions are not available for situations involving more than two samples when sample sizes are unequal (Conover 1971). Since $\underline{n}$ (the number of points which

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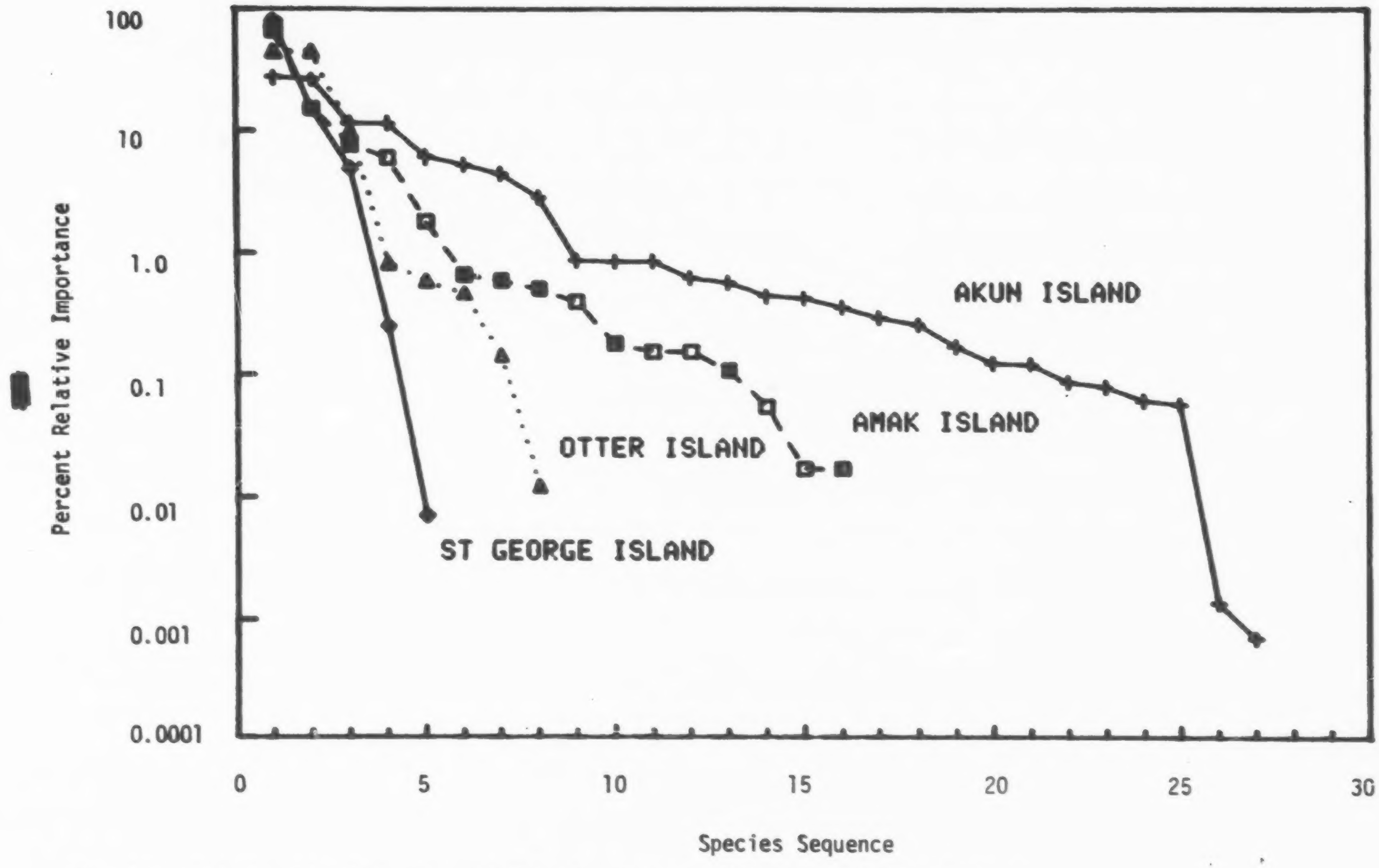

Fig. 84. Relationship between relative importance (biomass expressed as a percentage on a logarithmic scale) and rank of mollusks at four islands, the shores of two of which (Otter and St. George islands) are frequently scoured by sea ice.

determine the curve) for the species-abundance curve is the number of species in the collection and not the number of samples collected, one has no control over its size. Values of n were unequal among the four sites.

The Smirnov test of the empirical distribution functions of St. George Island vs. Akun Island was not significant ($T_1 = 0.326$, $N_1 = 5$, $N_2 = 27$). Presumably this holds for comparisons between islands with less divergent species-biomass distributions, e.g. between Otter Island and Akun Island or between St. George Island and Amak Island. This result is surprising because the curves appear markedly different when compared visually (Fig. 84). The test is conservative when the random variables are discrete, and use of the asymptotic approximation for large sample sizes further increases the conservatism. The Smirnov test may not be powerful enough to test the difference between species-importance curves when species-poor communities are involved. The marked difference in form in the species-importance curves for the Pribilof Islands versus those for Amak and Akun Islands suggests that there are real differences in the distribution of biomass among species between these two groups of islands, despite our inability to demonstrate this difference with the appropriate statistical test.

2. Effects on Community Dominants and Succession

Annual ice scouring of the intertidal region is a frequent and severe disturbance which repeatedly denudes the rocky substratum and would be expected to prevent benthic communities from developing beyond the early stages of succession. Under such conditions one would expect these communities to be composed of species which can colonize bare areas rapidly, i.e., fugitive species (Hutchinson 1951) or those which have a refuge during scouring episodes. Conversely, species which normally colonize during later stages of succession or those which have no refuge, such as sessile species, should be absent or low in abundance where the frequency of scouring is high.

The earliest colonizers of denuded surfaces are usually ephemeral algae such as diatoms, the filamentous green algae, Spongomorpha spp., foliose green (Ulvoid) and red (Porphyra) algae, etc. These species have good powers of dispersal but suffer in competition with other algae for space or light (Dayton 1975), or are preferentially grazed by herbivores (Lubchenco and Menge 1978), and therefore usually persist for no more than a few months.

Other fugitive algae (e.g., Halosaccion glandiforme) usually appear after the ephemeral species, and are, in turn, generally followed by a sequence of species with lower powers of dispersal and slower growth rates. Among this last group are species like Alaria, Fucus, Hedophyllum, and Laminaria, which grow to a large size, form a canopy over other members of the community, and apparently dominate other algae in competition for light and space (Dayton 1975). These species usually dominate communities where the frequency of disturbance is low.

Our results show that ephemeral algae (species were designated ephemeral after Appendix 1 of Lubchenco and Menge [1978]) rarely dominated intertidal plots at St. George and Otter Islands and never did so at Amak and Akun Islands (Table 36). The number of species of ephemeral algae was about the same at sites on Otter, Amak, and Akun Islands, but markedly lower (only *Porphyra* sp. was present) at the St. George Island site (Table 37). The filamentous green algae *Spongomorpha spinescens* and the ulvoids *Monostroma* sp., *M. fuscum*, and *Ulva lactuca* were absent from plots collected at the Pribilof Island sites, but present at Amak or Akun Islands (Table 37). Conversely, only *Spongomorpha* sp. and *S. arcta* were present at a Pribilof Island site (Otter Island) but absent at the Amak and Akun Island sites.

The organisms which most frequently dominated (had the greatest biomass in) plots at the Pribilof Island sites were *Halosaccion glandiforme* and *Halosaccion* sp. (We do not have conclusive evidence that these species are truly ecologically dominant over ephemeral algae in the sense that they preempt the greatest share of a limiting resource. It seems likely that, because they are larger than most ephemeral algae, *Halosaccion* spp. would be more successful in occupying space or intercepting light, two resources which are most likely to be limiting in intertidal systems. However, other mechanisms such as selective herbivory may cause the apparent dominance of *Halosaccion* in this system). *H. glandiforme* dominated half of the lower intertidal plots at St. George Island, all of the upper and, together with *Halosaccion* sp., most (66%) of the lower plots at Otter Island (Table 36). Neither species dominated any plot at Amak or Akun Islands, but *H. glandiforme* was present in 30-50% of all plots at both sites (except those in the upper intertidal zone at Akun Island) (Table 36, Fig. 85).

H. glandiforme is an annual. At Amchitka Island, Alaska (midway along the Aleutian Island chain) the spores of this species settle at all times of year except winter and the thalli grow rapidly (Lebednik and Palmisano 1977). Dayton (1975) classifies it as a fugitive species on the outer coast of Washington although apparently it can settle and grow in the understory of a "climax" community (Lebednik and Palmisano op. cit.). *H. glandiforme* can persist on the same site for several years (Lebednik and Palmisano op. cit.), therefore, its presence in the community does not necessarily indicate that the community is at an early stage of succession. However, it is unlikely that *H. glandiforme* could persistently dominate the community to the apparent exclusion of the canopy species (Table 36) unless some mechanism prevented the canopy species from settling among and growing over stands of *H. glandiforme*.

Three species of canopy-forming algae, *Fucus distichus*, *Alaria* sp., and *A. taeniata*, were present in plots in the Pribilof Islands (Table 38). None of these species dominated plots at St. George Island, although *Fucus* and *Alaria* sp. were present in the lower intertidal zone. At Otter Island, *Alaria* sp. or *A. taeniata* dominated some lower intertidal plots, but were highly variable in biomass (Tables 36 and 38).

The number of species, frequency of dominance, and average wet weight of canopy species tended to increase from the Pribilof Islands to Amak and Akun Islands. At Amak Island, *F. distichus* and *Alaria* sp. dominated (in biomass) plots in the upper intertidal zone (Table 36). These two species and *A. taeniata* dominated plots in the lower intertidal zone. Three additional canopy species, *Hedophyllum sessile*, *Laminaria* sp., and *L. longipes*, occurred in but never dominated lower intertidal plots at Akun Island (Table 38). *Fucus*, *Odonthalia floccosa*, and the barnacle *Balanus cariosus* had the

Table 36. Dominant (in biomass) species, frequency of dominance (D) expressed as the percentage of quadrats in which each species had the greatest wet weight, and frequency of occurrence (F) of each species in upper (upper number) and lower (lower number) intertidal zones at Amak, Akun, and the Pribilof Islands. All species showing the greatest wet weight in at least one quadrat/level/site combination are included. Dash means species was never dominant (D column) or was absent (F column) at the particular level and site.

Species[a]	St. George Island		Otter Island		Amak Island		Akun Island	
	D	F	D	F	D	F	D	F
Spongomorpha sp.	-	-	-	-	-	-	-	-
	-	-	6	.06	-	-	-	-
Porphyra sp.	-	-	-	-	-	.5	-	-
	25	.75	-	.06	-	.17	-	-
Halosaccion glandiforme	-	-	100	1.0	-	.5	-	-
	50	1.0	22	.78	-	.33	-	.4
Halosaccion sp.	-	-	-	.33	-	-	-	-
	-	-	44	.83	-	-	-	-
Alaria taeniata	-	-	-	-	-	-	-	-
	-	-	6	.06	17	.17	-	.2
Alaria sp.	-	-	-	-	50	.5	33	.33
	-	.25	28	.44	17	.5	-	.1
Odonthalia floccosa	-	-	-	-	-	.5	33	1.0
	-	-	-	-	-	.1	10	.8
Fucus distichus	-	-	-	-	50	1.0	33	1.0
	-	.75	-	-	67	1.0	30	.9
Littorina sitkana	-	.5	-	1.0	-	.5	-	.67
	25	1.0	-	.83	-	.33	-	.7
Littorina sp.	50[b]	.5	-	-	-	-	-	-
	-	-	-	-	-	-	-	-
Balanus cariosus	-	-	-	-	-	-	-	.67
	-	-	-	-	-	.33	60	.9

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a. Algae are listed roughly in order of increasing persistence in undisturbed environments.

b. Only two plots were sampled in the upper intertidal zone at St. George Island; one was bare.

Table 37. Ephemeral species[a] and mean wet weight (g) in upper (upper number) and lower (lower number) intertidal zones at Amak, Akun, and the Pribilof Islands. Dash means species was absent from the particular level and site. SD = standard deviation.

Species	St. George Island		Otter Island		Amak Island		Akun Island	
	$\bar{x}$	SD	$\bar{x}$	SD	$\bar{x}$	SD	$\bar{x}$	SD
Spongomorpha sp.	-	-	-	-	-	-	-	-
	-	-	.003	.01	-	-	-	-
S. arcta	-	-	-	-	-	-	-	-
	-	-	1.3	3.7	-	-	-	-
S. spinescens	-	-	-	-	1.0	1.4	-	-
	-	-	-	-	.008	.02	-	-
Monostroma sp.	-	-	-	-	-	-	-	-
	-	-	-	-	-	-	1.3	4.0
M. fuscum	-	-	-	-	-	-	-	-
	-	-	-	-	.01	.03	3.1	7.8
Ulva lactuca	-	-	-	-	-	-	-	-
	-	-	-	-	-	-	.2	.6
Pylaiella littoralis	-	-	-	-	-	-	-	-
	-	-	.01	.05	-	-	.4	.8
Scytosiphon lomentaria	-	-	-	-	-	-	-	-
	-	-	.3	.9	.6	1.4	-	-
Porphyra sp.	-	-	-	-	18.2	26.2	-	-
	9.1	15.8	.009	.04	15.2	37.2	-	-

a. Species of algae were designated ephemeral from Appendix 1 of Lubchenco and Menge (1978).

Table 38. Canopy species and mean wet weight ($\bar{x}$) in upper (upper number) and lower (lower number) intertidal zones at Amak, Akun, and the Pribilof Islands. Dash (--) means species was absent from the particular level and site.

Species[a]	St. George Island $\bar{x}$	St. George Island SD	Otter Island $\bar{x}$	Otter Island SD	Amak Island $\bar{x}$	Amak Island SD	Akun Island $\bar{x}$	Akun Island SD
Fucus distichus	--	--	--	--	44.8	57.4	362.5	409.8
	0.08	0.10	--	--	108.6	113.6	165.5	158.6
Alaria taeniata	--	--	--	--	--	--	--	--
	--	--	7.3	31.0	44.7	109.5	22.5	52.7
Alaria sp.	--	--	--	--	121.6	172.0	45.8	79.4
	3.6	7.2	87.4	272.4	53.3	130.1	2.5	8.0
Hedophyllum sessile	--	--	--	--	--	--	--	--
	--	--	--	--	--	--	16.1	42.2
Laminaria longipes	--	--	--	--	--	--	--	--
	--	--	--	--	--	--	7.4	16.4
Laminaria sp.	--	--	--	--	--	--	--	--
	--	--	--	--	--	--	0.09	0.3

a. Species were designated as canopy species (Dayton 1975, Menge 1976).

greatest wet weight in lower intertidal plots. *Fucus*, *Odonthalia*, and *Alaria* sp. dominated upper plots there.

3. Competition, Herbivory, and Community Development after Ice-scouring

Each episode of ice-scouring sets back the process of succession to an early stage by creating bare rock which ephemeral species can colonize. Our data show that at 4 months after the last scouring episode, *Spongomorpha* sp. and *S. arcta* were the only ephemeral algae with greater biomass on scoured than on unscoured islands (Table 37) and that *Halosaccion* spp. dominated most plots on scoured islands.

Two mechanisms acting alone or in concert may have allowed *Halosaccion* spp. to dominate plots on scoured shores. The first is that *Halosaccion* settles concurrently with or after the early-colonizing ephemeral species and simply outcompetes them for space or light. The second involves selective herbivory by snails on ephemeral algae; by preferentially grazing ephemeral species the snails allow *Halosaccion* to settle and grow in the absence of competition by the ephemeral species. Lubchenco and Menge (1978) have shown that grazing by *Littorina littorea* on ephemeral algae accelerates the development of *Chondrus crispus* (Irish moss) beds in New England.

Unambiguous evaluation of the relative importance of each of these mechanisms requires experimental manipulation of herbivore and *Halosaccion* populations, but examination of the relationship of the abundance of ephemeral algae and *Halosaccion* on scoured and unscoured surfaces to relative intensity of herbivory may shed light on the mechanism resulting in dominance of *Halosaccion* at St. George and Otter Islands. We assume that the intensity of herbivory increases proportionately with herbivore biomass.

The most abundant intertidal grazer in plots in the intertidal zone at our Pribilof Island sites was *Littorina sitkana* (Fig. 86). Three other molluscan herbivores, *Haloconcha reflexa*, *Margarites helicinus*, and *Schizoplax brandtii* were present in lower intertidal plots at Otter Island, but individuals were smaller and less abundant than those of *Littorina*. One *Katharina tunicata*

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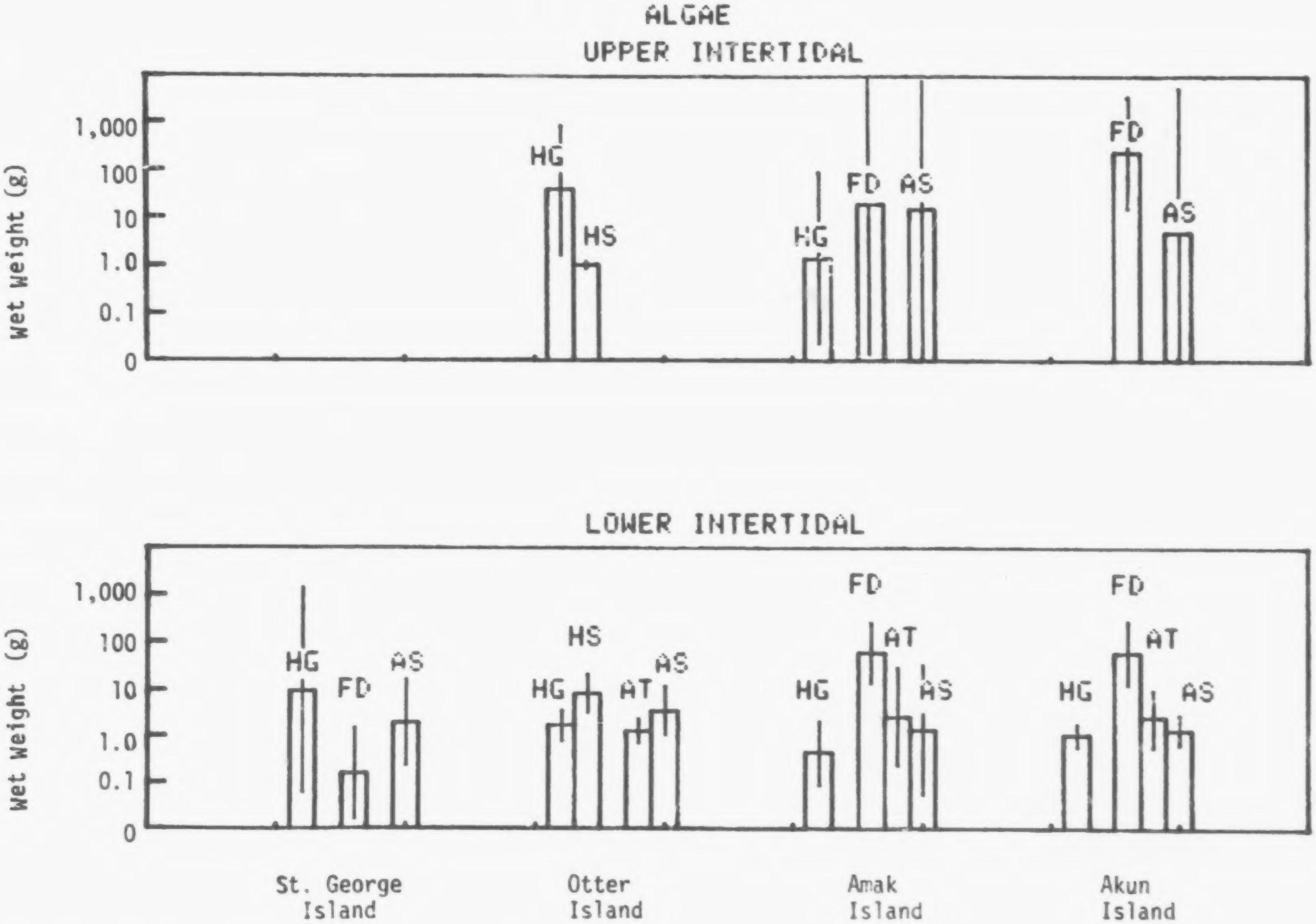

Fig. 85. Mean weight of four species of algae at four islands in the Bering Sea. AS, Alaria sp.; AT, Alaria taeniata; FD, Fucus distichus; HG, Halosaccion glandiforme; HS, Halosaccion sp. Vertical lines are 95% confidence intervals.

was found in a lower intertidal plot at St. George Island. *L. sitkana* and *L. aleutica* were common in plots at Amak and Akun Islands (Fig. 86).

Little is known about the food habits of *Littorina sitkana*; nothing is known of those of *L. aleutica*. Caged *L. sitkana* will graze diatoms and probably also the sporelings of *Ulva* and *Enteromorpha* (Behrens 1971). Other species of *Littorina* eat mainly diatoms (but see Berry 1961 and Hayes 1929) and small, tender algae (Table 39) most of which are ephemeral (e.g., Lubchenco 1978). However, *L. scutulata* and *L. littorea* will feed on large plants (Dahl 1964, Bakker 1959, Hayes 1929, Lubchenco 1978).

Other grazers which could reduce the abundance of ephemeral algae are limpets. Four species of limpets, *Collisella* sp., *C. pelta*, *Notoacmea scutum*, and *N. persona*, occurred at our intertidal study sites (MHW to MLW) on Amak and Akun Islands. Limpets were absent from the intertidal plots at St. George and Otter Islands, but *C. pelta* occurred in 70% (*n* = 34) of the plots below MLW at St. George Island.

Notoacmea scutum, *C. pelta*, and other eastern Pacific limpets of the genus *Collisella* are known to graze diatoms, blue green algae, and other microscopic algae (Castenholz 1961, Haven 1973, Nicotri 1977), but their diets probably include a greater proportion of macroscopic algae than does that of *Littorina*. Craig (1968) found that *Acmaea pelta* (= *Collisella pelta*) ingests a wide variety of microscopic and macroscopic algae including small fragments of large plants such as *Pelvetia* and *Egregia*. Walker (1968) concluded from a study of the configuration of the gut of *Acmaea scutum* (= *Notoacmea scutum*) that it probably feeds mainly on larger algae; she found fragments of flat encrusting algae in its gut. We could find no information on the diet of *N. persona*.

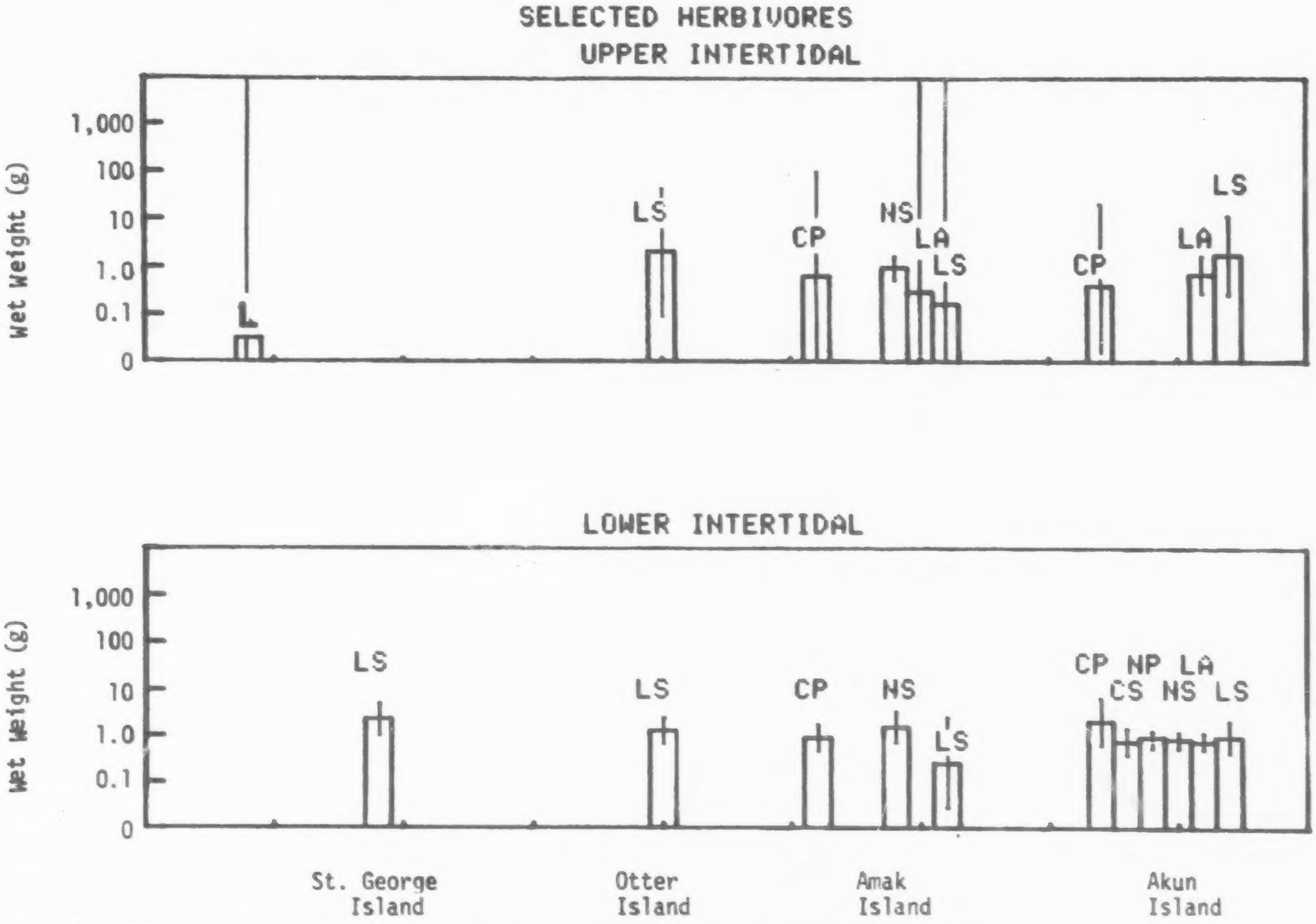

Fig. 86. Mean weight of seven species of herbivore on ice-scoured (St. George and Otter Islands) and unscoured islands in the Bering Sea. CP, Collisella pelta; CS, Collisella sp.; L, Littorina sp.; LA, Littorina aleutica; LS, Littorina sitkana; NP, Notoacmea persona; NS, Notoacmea scutum. Vertical lines are 95% confidence intervals.

Table 39. Food of *Littorina*[a]

Food	North Pacific Species	
	Littorina scutulata	*Littorina planaxis*
Bacillariophyceae		
Diatoms	2,5,7	5,7
Cyanophyceae		
Unicellular blue green algae	5,7	5,7
Dermocarpa	5	5
Spirulina	5	5
Calothrix	5	5
Plectonema	5	5
Chlorophyceae		
Prasiola		4[d]
Ulva	4	
Spongomorpha	5	5
Cladophora	4	4
Phaeophyceae		
Laminaria	4	
Pelvetia	4	
Rhodophyceae		
Porphyra	4	4[e]
Rhodochorton	5	5
Rhodoglossum		4[e]
Endocladia	5	4[e],5

Food	North Atlantic Species	
	Littorina littorea	*Littorina saxatilis*
Bacillariophyceae		
Diatoms	3,8[b]	1[b]
Chlorophyceae		
Ulothrix-Urospora	9[c]	
Monostroma	8,9	
Enteromorpha	3,9	
Ulva	3,9	
Spongomorpha	9	
Cladophora	8,9	
Pseudoendoclonium		1
Phaeophyceae		
Ectocarpus-Pylaiella	8,9	
Elachistea	9	
Ascophyllum	6,8,9	
Fucus	6,8,9	
Pelvetia	6	
Petalonia	9	
Scytosiphon	9	
Rhodophyceae		
Porphyra	9	
Rhodymenia	8,9	
Ceramium	9	
Halosaccion	9	

Table 39. Continued.

a. Numbers in body of table refer to the following papers: 1, Berry (1961); 2, Castenholz (1961); 3, Newell (1958); 4, Dahl (1964); 5, Foster (1964); 6, Bakker (1959); 7, Glynn (1965); 8, Hayes (1929); 9, Lubchenco (1978). Several additional references on the food habits of Littorina were unavailable to me at the time of this writing (see Pettitt 1974).
b. Diatoms in gut did not appear to be digested.
c. All genera from Lubchenco's list of highly preferred food are included here. Not all genera of medium and low preference ranking are included. See Table 1 of Lubchenco (1978) for complete list.
d. Small plants eaten.
e. Finely chopped but not whole plants eaten.

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At the time of this study the intensity of herbivory on ephemeral algae was apparently no greater at Otter and St. George Islands than at Amak and Akun Islands. A one-way anova (upper and lower intertidal zones combined) revealed that the biomass of Littorina spp. was significantly greater at the St. George and Otter Island sites combined than at the Akun and Amak sites combined; a posteriori comparisons (Scheffé's [1953] test) of Otter and St. George Islands with Amak and Akun Islands separately revealed that the lower biomass of Littorina spp. at the Amak site accounted for the difference (Table 40). There was no significant difference between the average biomass of Littorina at St. George and Otter Island and that at Akun Island.

The absence of limpets on our plots at the Pribilof Islands suggested that there were fewer species of known consumers of ephemeral algae on scoured surfaces there. Haloconcha reflexa, Margarites helicinus, and Schizoplax brandtii may ingest young sporophytes and gametophytes of ephemeral algae while grazing, but because they were smaller and less numerous than L. sitkana on St. George and Otter Islands, they probably cannot control populations of ephemeral algae there.

Similar biomass levels of Littorina at the Pribilof Islands and Akun Island may be misleading. Greater species richness and weight of large macrophytes at Akun Island (Table 38) results in greater spatial heterogeneity and probably also greater effective grazing area for Littorina. Therefore, the biomass of Littorina per unit effective grazing area may be much greater on frequently scoured rock at the Pribilof Islands. We have no measure of the effective grazing area contributed by macrophytes at Akun Island.

Finally, the data may reflect the results of herbivory at a time when the intensity of herbivory is reduced, after the period when colonizing algae are settling and growing most rapidly and consumers are exerting their greatest

Table 40. Tests of significance of *Littorina* wet weights (*L. sitkana* and *L. aleutica* combined) in the rocky intertidal region at the Pribilof Islands compared with Amak and Akun Islands. ** = test significant.

Source	d.f.	Anova S.S.	M.S.	F
Site	3	143.8	47.9	5.36**
Error	44	393.5	8.9	
Total	47	537.4		

Treatment	n	Mean of transformed counts[a]	95% Confidence interval Lower limit	95% Confidence interval Upper limit
St. George Island (G)	6	5.27	2.81	7.73
Otter Island (O)	21	6.24	4.93	7.56
Amak Island (Am)	8	1.31	-0.82	3.44
Akun Island (Ak)	13	5.43	3.75	7.1

Comparison	S	K	n_0	F	Significance
G-0 vs Am-Ak	-	-	-	6.14	$p < 0.05$[b]
G-O vs Am	8.94	4	44	--	$p < 0.05$[c]
G-O vs Ak	8.94	4	44	--	n.s.[c]

a. Weights were scaled (x 1000) prior to transformation (log [x + 1]) to avoid negative characteristics.

b. *A priori* orthogonal comparison.

c. *A posteriori* comparison with Scheffé's test. S, K, and n_0 are statistics of Scheffé's test. S^2 = the experimentwise error, K = number of cell means, n_0 = degrees of freedom of the error term of the anova.

effect. An evaluation of the role of herbivores in community development on rock surfaces scoured by ice awaits further study. The populations of plants and herbivores in this system are probably amenable to experimental manipulation.

4. Populations of Sessile Invertebrates

We would expect sessile invertebrates to be absent or low in abundance (or weight) where the frequency of scouring is high; such organisms cannot retreat under or into crevices in rocks during scouring episodes and most can recolonize scoured rock only by settlement of planktonic larvae. Where these species have colonized scoured surfaces, they should be represented solely by young individuals which have settled since the last scouring event.

Our data tend to support the above suppositions. Four species, *Mytilus edulis*, *Chthamalus dalli*, *Balanus glandula*, and *B. cariosus*, were chosen for study because they are widespread in Alaska and where they occur they usually occupy greater proportions of rocky intertidal space than other sessile invertebrates. None of these species was present in plots in the upper intertidal zone in the Pribilof Islands (Fig. 87). (However, small *Mytilus* were collected in three of four and two of seven plots above MHW [and therefore above the upper tidal level considered in the present report] at St. George and Otter Islands, respectively.) *Mytilus* was present in lower intertidal plots at both St. George and Otter Islands, but was represented only by small individuals ($\leq$15 mm in length) which were highly variable in biomass among plots (Fig. 87).

Balanus glandula was represented by a single individual in one plot in the lower intertidal zone at Otter Island. *B. cariosus* was absent from plots in both intertidal zones in the Pribilof Islands, but was present in two of 27 plots below mean low water (MLW) at St. George Island. Unidentified barnacles (*Balanus* sp.) were collected in three of the 27 plots.

Barnacles tended to be more abundant at Amak and Akun Islands than at Otter and St. George Islands (Fig. 87). *Balanus glandula* was absent from upper and lower intertidal zones at Akun and Amak Islands respectively, but

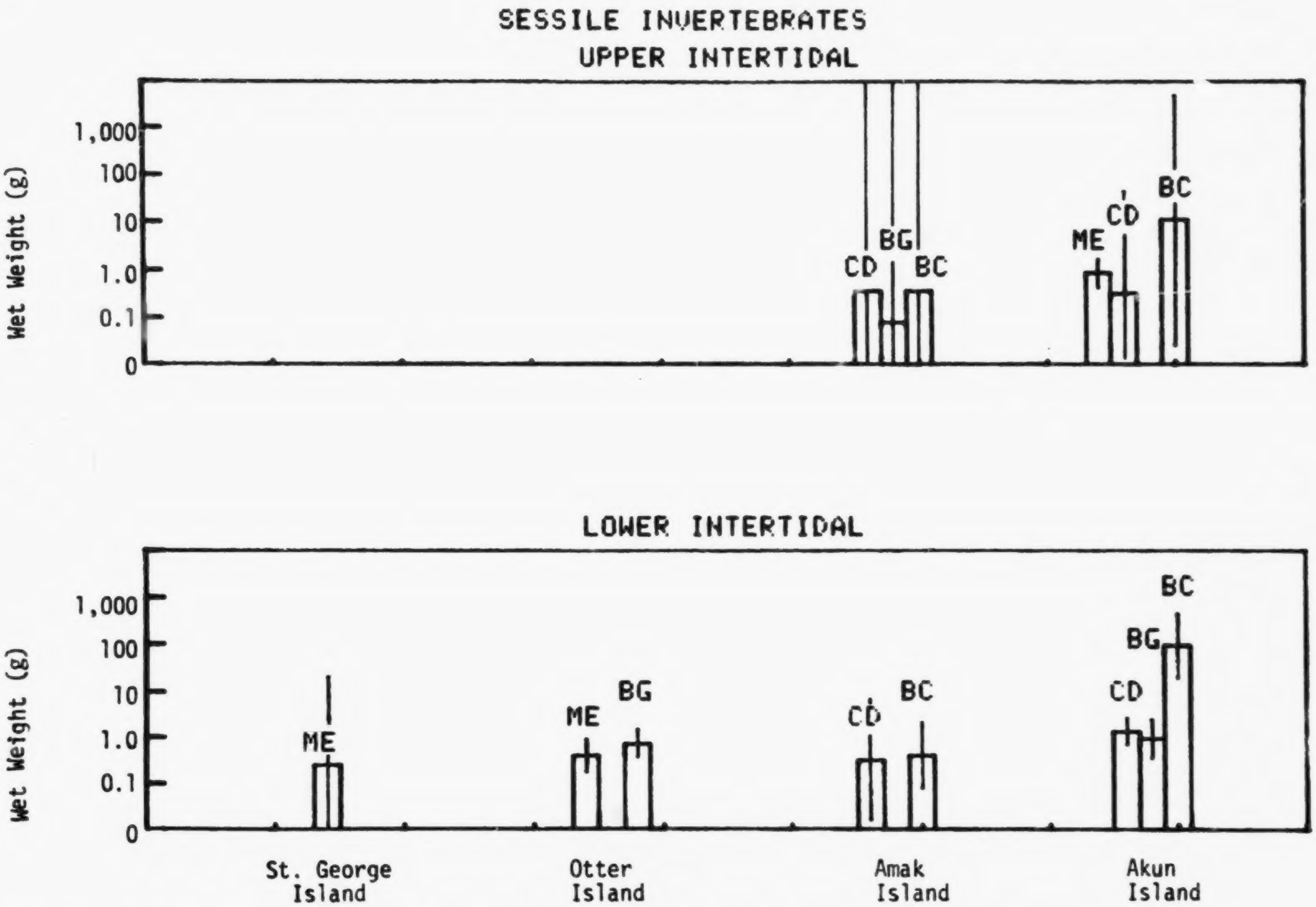

Fig. 87. Mean weight of sessile invertebrates at two ice-scoured (St. George and Otter Islands) and two unscoured islands in the Bering Sea. BC, *Balanus cariosus*; BG, *Balanus glandula*; CD, *Chthamalus dalli*; ME, *Mytilus edulis*. Vertical lines are 95% confidence intervals.

was present above (in two of two plots) and below (in one of 11 plots) the respective upper and lower limits of this study at these two sites.

_M_. _edulis_ appears to be an exception to the trend toward greater abundance among major sessile animals with decreasing frequency of ice scouring. Although three large ($\geq$20 mm) _Mytilus_ were present in one plot located below MLW at Akun Island, only a few (eight) small individuals were found in one plot (of five) in the upper intertidal zone there. _M_. _edulis_ was absent from all plots sampled at Amak Island. Based on the data at hand we are unable to account for the apparently small populations of _M_. _edulis_ at Amak and Akun Islands.

5. Refuges from Ice Scouring

Our quantitative sampling emphasized upper rock surfaces at all sites. The effects of scouring by ice are most pronounced on these surfaces (Fig. 88). As shown above, the species richness of marine organisms on these surfaces is low. However, our observations and photographs of the biota between and beneath closely packed boulders and in rock crevices indicate that species richness in these places is more comparable to that of unscoured surfaces at Amak and Akun Islands (T. R. Merrell, Jr., 1979, pers. commun., Manager, Environmental Program, Northwest and Alaska Fisheries Center Auke Bay Laboratory, Auke Bay, Alaska 99821). Such protected places apparently are refugia from scouring for those species whose growth form and light requirements permit them to occupy such microhabitats.

We also noted one area of shore near our transect site at Garden Cove, St. George Island (Fig. 89), where deep fissures in the bedrock and offshore reefs protected much of the rock surface from ice scouring. One of us (R.M.) made counts of organisms there; the biota included large Collisella pelta, urchins, and several species of sessile organisms including B. cariosus, two sponges, hydroids, bryozoans, tubeworms, tunicates and coralline algae.

Subtidal observations by divers indicated that the effect of ice-scour generally reached 3-4 m in depth. Above this lower limit only newly-settled algae occupied the upper surfaces of rock. Larger plants, presumably survivors of one or more winters, were found only in crevices. The fronds of these plants were sharply cropped at the level of the rock surface. Below 4 m, large perennial plants such as Laminaria and Thalassiophyllum flourished. Several-year-old Constantinea plants were observed at 5 m.

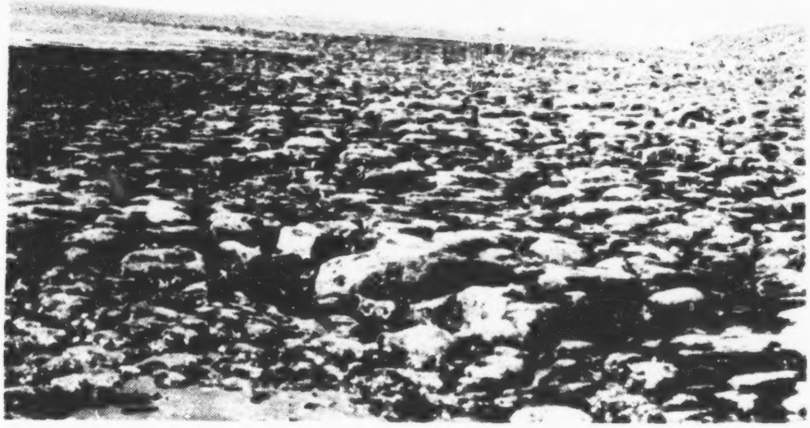

Fig. 88. Intertidal region at English Bay, St. Paul Island. Note lack of biological cover on tops of boulders. 10 June 1976.

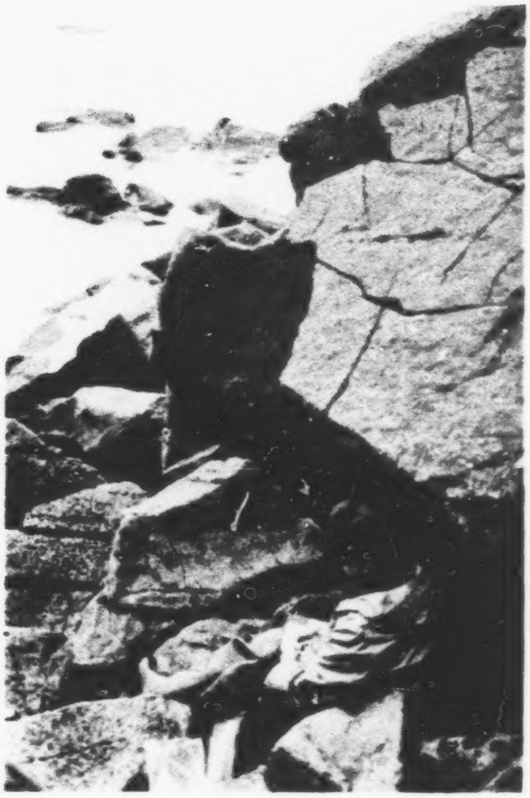

Fig. 89. Garden Cove site showing the deep fissures in and crevices between rocks. These cracks serve as refuges for intertidal organisms.

Below the depth of influence by sea ice, the sublittoral region appeared exceptionally rich in species when compared with the intertidal region.

In the Pribilof Islands, greater species richness among invertebrates and algae in refugia in the intertidal region and below the lower limit of scouring by sea ice in the sublittoral region suggests that the relative effect of island size and distance from the nearest source area as compared with that of scouring by ice is probably small. We do not have quantitative data on species richness in the refugia; such data would be helpful for assessing the role of island biogeographic effects among our study sites.

VIII. DISCUSSION

Scouring by sea ice is probably the most important disturbance affecting intertidal community structure at the Pribilof Islands. Our results indicate that species diversity is low where the frequency of scouring is high and that the relationship is probably causative. However, Huston (1979) contends that the frequency of disturbance alone cannot adequately predict species diversity, and that diversity represents a dynamic equilibrium between the rate of population reduction by disturbance or predation and the rate of approach to competitive equilibrium. Thus in order to fully evaluate the role of sea ice on an intertidal community, we need to know the rates of growth of the populations of potential competitors.

Information on both of these parameters is important to the study of the impact of oil pollution on an intertidal community because an oil spill could act both to increase density-independent mortality in many species and to limit the population growth rates of surviving competitors by supressing primary production, interfering with feeding behavior, or reducing fecundity or setting success. These effects could offset one another if the rate of competitive displacement and the frequency of reduction are not markedly disparate (i.e., that one is not of overriding importance) and the oil spill is mild. However, in ice-stressed systems the community is sufficiently far from competitive equilibrium owing to the high rate of disturbance by sea ice that species diversity would more likely be further reduced.

The most important characteristic of ice-stressed coasts that allows species to remain in the system is the availability of refuges from ice scouring. Woodin (1978) suggests five major categories of refuges from disturbance: 1) temporal periods outside the activity range of the disturbance process, 2) temporal periods within the activity range of the disturbance process, 3) spatial zones beyond the activity range of the disturbance process, 4) physical heterogeneities within the activity range of the disturbance process, and 5) biologically generated refuges within the activity range of the disturbance process.

Since it is unlikely that biogenic structures could withstand ice scour, category 5 is probably unimportant in ice-stressed systems. Some motile species might be able to migrate onto scoured surfaces during a scouring episode, for example when the ice is temporarily lifted from lower intertidal surfaces at high tide (category 2), but there seems to be little advantage for an individual to do this since it must return to a spatial refuge within 12 h at most.

Spatial zones beyond the range of scouring (e.g., supra- or sublittoral habitats) could be important for some species. However, since the supra-littoral zone is above the upper physiological limits of most intertidal species and they are often prevented from establishing populations in the sublittoral region by the activities of predators and competitors (Connell 1972), few of them could occupy these habitats.

Temporal periods outside of (i.e., June through December) and physical heterogeneities within (interstices of boulder fields and crevices) the activity range of ice scouring remain the primary refuges available to intertidal organisms at the Pribilof Islands. Of these, the latter category is probably more important because a species using the temporal refuge would require a planktonic stage (e.g., spores, gametes, or larvae) which could weather the scouring episode and would thereby risk having its propagules swept away from the islands whereas refuges provided by spatial heterogeneities could harbor many life stages (e.g., minute sporophytes, microscopic gametophytes, and in some cases macroscopic algae and egg masses, juveniles, and adults of many invertebrates.

None of the above-mentioned refuges from ice scouring could offer complete protection from an oil spill. Depending on wind and wave action, both supra- and sublittoral habitats could be contaminated. An oil spill reaching the shores of the Pribilof Islands in June through December would immediately interrupt the progress of ecological succession. Although an offshore oil spill might be temporarily prevented from coming ashore during the period when the Pribilofs are iced in, depending on wind and currents, oil could reach the shore as the ice gradually recedes north. Since oil in contact with ice weathers very slowly (e.g., Atlas et al. 1978), the toxicity of the oil would probably still be great when it eventually reached the shore. Oil reaching the shore when ice is present could be abraded, redistributed, and dispersed somewhat by ice scouring, but sea ice also prevents wave generation and dampens existing waves thereby reducing mechanical dispersal which could be the most important dispersing agent at higher latitudes (Owens 1978).

Physical heterogeneities on rocky shores in the Pribilof Islands are coarse-grained (compared with gravelly, sandy, or muddy beaches), interstitial spaces are large; therefore, even heavy oils can penetrate them. Moreover, rates of abrasion and dispersion are reduced in coarse-grained substrates (Owens 1978). The primary refuge from ice-scour for intertidal organisms would therefore become rapidly contaminated for a prolonged period if an oil spill washed ashore at the Pribilof Islands. The net effect of a spill probably would be a proportionately large reduction in species richness involving local extinction in some cases and a prolonged period of return to a natural community.

VIII. CONCLUSIONS

1. Comparisons of intertidal communities at islands whose shores had recently been subject to scouring by sea ice with those at unscoured islands indicate that ice scouring significantly reduces species richness. (Reasonably firm).

2. The distribution of biomass among species tends towards a greater concentration of dominance among a few species on shores recently scoured by ice. (Tentative).

3. By midsummer, 4 months after the end of a scouring event, the intertidal community on shores scoured by ice is still dominated by fugitive and ephemeral species whereas canopy species dominate unscoured shores. (Preliminary).

4. Major competitors for space in the mid-intertidal zone (these competitors are generally sessile invertebrates) tend to be absent from or small and low in abundance on unprotected surfaces where the frequency of scouring is high. (Preliminary).

IX. NEEDS FOR FURTHER STUDY

Our conclusions need to be tested experimentally. Although scouring by ice seems to be the major structuring force in intertidal communities at the Pribilof Islands we need to examine more fully the role of predation, especially herbivory, and competition in ecological succession after a scouring episode. It should be possible to protect areas of exposed rock by artificial shelters made of steel reinforcing bars or concrete and thereby provide experimental refuges wherein species densities could be manipulated and changes in community structure could be followed through winter and spring in the absence of scouring.

Natural refuges should be studied to learn what intrinsic and extrinsic factors determine which species can use them and how they use them.

Rates of competitive displacement should be estimated in order to quantify the dependence of species diversity on this factor in combination with the frequency of ice scouring.

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APPENDICES

APPENDIX I

Key to the species and groups identified in Eider Point samples[a].

1. *Balanus cariosus*
2. Alariaceae cf. *Alaria crispa*
3. *Strongylocentrotus droebachiensis*
4. *Alaria crispa taeniata*
5. *Katharina tunicata*
6. *Hiatella arctica*
7. *Palmaria palmata*
8. *Mytilus edulis*
9. *Spongomorpha spinescens*
10. *Leptasterias* sp.
11. Phaeophyta
12. *Rhodymenia* sp.
13. Synoicidae
14. *Collisella pelta*
15. *Notoacmea scutum*
16. Zoantharia Actiniaria
    Nynantheae thenaria
17. Tunicate frag.
18. *Nucella canaliculata*
19. *Pterosiphonia bipinnata*
20. *Leptasterias leptodoma*
21. *Monostroma fuscum*
22. *Cirratulus cirratus*
23. *Mopalia ciliata*
24. *Collisella* sp.
25. *Iridaea* sp.
26. *Cancer oregonensis*
27. *Analipus japonicus*
28. *Nereis pelagica*
29. *Nereis vexillosa*
30. *Amphiglena* sp.
31. *Molgula oregonia*
32. *Cancer* sp.
33. *Nicolea zostericola*
34. *Cucumaria pseudocurata*
35. *Leptasterias camtschatica*
36. Calcareous red alga
37. *Colpomenia bullosa*
38. Porifera
39. Rhynchocoela
40. *Scrupocellaria arctica*
41. *Idotea wosnesenskii*
42. *Capitella capitata*
43. *Dynamenella sheareri*
44. *Leptasterias hexactis*
45. *Polycirrus medusa*
46. *Odonthalia floccosa*
47. *Tonicella lineata*
48. *Mopalia* sp.
49. *Spongomorpha* sp.
50. *Antithamnion* sp.
51. *Eulalia quadrioculata*
52. *Parallorchestes ochotensis*
55. Polychaete (frag.)
56. *Asabellides sibirica*
58. *Eteone longa*
59. *Typosyllis* sp.
60. Oligocheata
65. *Naineris quadricuspida*
66. *Exogone gemmifera*
67. Phyllodocidae
68. Eucarida Decapoda Pleacyemata
    Stenopodidea
70. *Potamilla neglecta*
76. *Typosyllis variegata*
77. Cnidaria
78. Nematoda
79. *Margarites beringensis*
87. *Pterosiphonia* sp.
88. Rhodophyta
89. Diptera larvae
90. *Parapleustes nautilis*
92. *Autolytus* sp.
94. *Munna chromatocephala*
96. *Chone gracilis*
97. Terebellidae
101. *Monostroma* sp.
104. Egg case
106. *Ammothea pribilofensis*
109. *Musculus olivaceous*
131. Chlorophyta
137. Gastropoda
138. *Parapleustes* sp.
139. *Paleonotis bellis*
148. *Leptasterias camtschatica*
151. *Balanus glandula*
154. *Achelia chelata*
157. *Littorina sitkana*
170. *Fabricia crenicollis*
174. *Henricia leviuscula*
175. *Schizobranchia insignis*
176. *Spongomorpha* cf. *mertensii*
177. *Ischyrocerus* cf. *tsvetkovae*
178. *Exosphaeroma amplicauda*
179. Echinodermata

[a] Not all species analyzed in the original data are presented in Figures 30 and 31; this accounts for periodic non-consecutive numbering in the table.

APPENDIX II

Species Lists

The nomenclature used in the following species lists generally follows that of the National Oceanographic Data Center (NODC) Taxonomic Code (Anonymous 1978) prepared by the Institute of Marine Science of the University of Alaska. The taxonomic order follows the code order of Anonymous (1978). Only live organisms in samples collected at our sampling sites are included. Usually only the phylum and class to which each species belongs are included in the lists unless one or more representatives of the phylum were not identified more specifically than to some higher taxon other than phylum or class (e.g. subclass, order, etc.) in which case the names of all other groups at that taxonomic level are included to avoid confusion over the relationships of other species in the phylum. The names of families are included especially if the group is diverse and organisms are frequently identified only to family within the group.

Only the presence of species are noted by an "x" unless an organism was not identified more specifically than to some higher taxonomic level. Several species in Appendix IID were collected from the subtidal region. They are noted by an asterisk after their names.

Appendix Table IIA.--List of the species of benthic plants and invertebrates at intertidal sites in the western Gulf of Alaska and northern Bristol Bay.

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
PHYLUM CHLOROPHYTA				
Family Ulotrichaceae				
Ulothrix sp.			x	
Family Monostromataceae				
Monostroma fuscum	x		x	x
Family Ulvaceae				
Ulva lactuca			x	x
Ulva sp.				x
Ulva fenestrata				x
Enteromorpha intestinalis	x		x	
Family Acrosiphoniaceae				
Spongomorpha arcta			x	
Spongomorpha sp.			x	
Spongomorpha spinescens			x	
Urospora mirabilis	x	x		x
Family Cladophoraceae				
Lola lubrica	x			
Cladophora sp.			x	
Cladophora seriacea	x			x
Rhizoclonium implexum	x			
PHYLUM CRYSOPHYTA				
Subphylum Bacillariophyceae	x		x	
PHYLUM PHAEOPHYTA				
Order Ectocarpales				
Family Ectocarpaceae				
Pylaiella littoralis	x	x	x	x
Ectocarpus simulans			x	x
Family Elachistaceae				
Elachistea fucicola	x			x

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Chordariaceae				
Analipus japonicus			x	
Order Dictyosiphonales				
Family Punctariaceae				
Melanosiphon intestinalis	x		x	
Soranthera ulvoidea			x	x
Family Sphacelariaceae				
Sphacelaria sp.	x	x		
Family Laminariaceae				
Laminaria sp.			x	x
Laminaria longipes				x
Family Alariaceae				
Alaria sp.			x	x
Alaria taeniata			x	x
Desmarestia viridis			x	
Desmarestia aculeata			x	x
Family Fucaceae				
Fucus sp.			x	
Fucus spiralis	x	x	x	x
Order Scytosiphonales				
Family Scytosiphonaceae				
Petalonia fascia			x	
Petalonia sp.				x
Scytosiphon lomentaria	x	x	x	x
PHYLUM RHODOPHYTA				
Family Bangiaceae				
Bangia fuscopurpurea			x	
Porphyra sp.			x	x

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Acrochaetiaceae				
Acrochaetium sp.				x
Family Cruoriaceae				
Petrocelis franciscana				x
Petrocelis middendorffii				x
Family Phyllophoraceae				
Ahnfeltia plicata		x		
Family Gigartinaceae				
Iridaea sp.				x
Iridaea cornucopiae			x	x
Gigartina pacifica				
Gigartina papillata				x
Gigartina stellata				x
Gigartina sp.				x
Order Cryptonemiales				
Family Dumontiaceae				
Cryptosiphonia woodii			x	x
Family Endocladiaceae				
Endocladia muricata			x	x
Gloiopeltis furcata				x
Family Corallinaceae				
Corallina sp.		x		x
Corallina vancouveriensis				x
Bossiella sp.				x
Bossiella chiloensis			x	x
Bossiella plumosa				x
Family Kalymeniaceae				
Callophyllis sp.				x
Callophyllis flabellulata			x	x
Callophyllis pinnata				x

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Rhodymeniaceae				
Rhodymenia palmata			x	x
Palmaria palmata			x	x
Halosaccion glandiforme	x		x	x
Family Ceramiaceae				
Microcladia borealis				x
Ptilota sp.				x
Neoptilota sp.				x
Neoptilota asplenoides				x
Family Rhodomelaceae				
Pterosiphonia bipinnata		x	x	x
Odonthalia floccosa		x	x	x
Odonthalia washingtoniensis				x
PHYLUM ANTHOPHYTA				
Zostera marina	x	x	x	
PHYLUM PORIFERA			x	x
PHYLUM CNIDARIA				
Order Hydroida				x
Obelia sp.				x
Class Anthozoa		x	x	x
PHYLUM PLATYHELMINTHES				
Class Turbellaria		x	x	x
PHYLUM RHYNCHOCOELA(Nemertea)		x	x	x
Emplectonema gracile			x	x
PHYLUM NEMATODA	x		x	x

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
PHYLUM ANNELIDA				
Class Polychaeta			x	x
Family Polynoidae			x	
Harmothoe imbricata			x	
Family Sigalionidae				
Pholoe minuta		x	x	
Family Chrysopetalidae				
Dysponetus sp.				x
Family Phyllodocidae				
Anaitides maculata		x		
Eteone longa		x	x	x
Eulalia quadrioculata			x	x
Eulalia bilineata			x	x
Family Syllidae				
Autolytus prismaticus			x	
Typosyllis sp.			x	x
Typosyllis elongata				x
Typosyllis stewarti			x	
Typosyllis fasciata			x	x
Typosyllis alternata			x	
Typosyllis pulchra			x	x
Typosyllis a. adamantea			x	x
Eusyllis sp.			x	
Exogone gemmifera		x		
Sphaerosyllis sp.			x	x
Sphaerosyllis pirifera				x
Syllides japonica			x	
Family Nereidae				
Nereis sp.		x	x	x
Nereis pelagica		x	x	x
Nereis vexillosa		x	x	
Nereis procera			x	

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Sphaerodoridae				
Sphaerodoropsis minuta				X
Family Goniadidae				
Glycinde picta			X	
Family Lumbrineridae				
Lumbrineris inflata			X	X
Family Dorvilleidae				
Protodorvillea sp.			X	
Family Orbiniidae				
Naineris quadricuspida			X	
Naineris laevigata			X	
Naineris sp.			X	
Family Spionidae				X
Polydora sp.			X	
Family Cirratulidae				
Cirratulus cirratus			X	X
Tharyx sp.			X	X
Family Capitellidae				
Capitella capitata		X	X	X
Family Terebellidae				
Polycirrus medusa			X	X
Family Sabellidae				
Chone gracilis				X
Potamilla neglecta			X	
Pseudopotamilla reniformis				X
Schizobranchia insignis			X	
Amphiglena pacifica			X	X
Fabricia sabella		X	X	X
Fabricia sp.			X	
Fabricia pacifica				X

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Sabellidae - cont'd.				
Fabricia crenicolis			x	x
Family Serpulidae				
Dexiospira spirillum			x	x
Class Oligochaeta				x
Family Enchytraeidae	x	x	x	x
PHYLUM MOLLUSCA				
Class Polyplacophora				
Schizoplax brandtii			x	x
Katharina tunicata			x	x
Class Bivalvia				
Mytilus edulis	x	x	x	x
Musculus discors		x		x
Musculus vernicosus				x
Musculus olivaceous				x
Musculus sp.			x	x
Modiolus modiolus	x	x	x	x
Pododesmus macroschisma				x
Turtonia minuta				x
Turtonia occidentalis		x	x	x
Protothaca staminea			x	
Hiatella arctica		x	x	x
Class Gastropoda				
Collisella sp.		x	x	x
Collisella pelta		x	x	x
Collisella digitalis			x	x
Notoacmea scutum		x		x
Notoacmea persona			x	x
Margarites sp.			x	x
Margarites pupillus			x	

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Class Gastropoda - cont'd.				
Margarites beringensis			x	x
Margarites helicinus			x	x
Moellaria costulata				x
Halochoncha reflexa				x
Lacuna sp.			x	
Lacuna marmorata			x	x
Lacuna vincta		x		
Littorina scutulata				x
Littorina sitkana	x	x	x	x
Littorina aleutica			x	x
Alvinia aurivillii				x
Barleeia subtenuis			x	
Barleeia sp.				x
Cerithiopsis sp.				x
Cerithiopsis stejnegeri			x	x
Melanella randolfi			x	
Nucella sp.			x	
Nucella canaliculata			x	
Nucella lamellosa			x	x
Nucella lima		x	x	
Buccinum baeri			x	x
Buccinum sp.			x	
Odostomia sp.				x
Cylichna sp.				x
Siphonaria thersites				x
PHYLUM ARTHROPODA				
Class Pseudoscorpionida				x
Class Acarina		x	x	x
Family Halicaridae	x			x

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Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Class Pycnogonida				
Ammothea latifrons				x
Ammothea pribilofensis				x
Class Crustacea				
Subclass Platycopa				x
Subclass Copepoda				
Order Harpacticoida				x
Subclass Cirripedia				
Order Thoracica				
Chthamalus dalli	x	x	x	x
Chthamalus sp.	x			
Balanus nubilis		x		
Balanus balanus		x		
Balanus cariosus			x	x
Balanus glandula	x	x	x	x
Balanus balanoides	x		x	
Balanus sp.	x		x	x
Order Cumacea				
Cumella sp.		x		
Order Tanaidacea		x		
Order Isopoda				
Family Sphaeromatidae				
Gnorimosphaeroma oregonensis			x	
Exosphaeroma amplicauda				x
Exosphaeroma sp.				x
Dynamenella sheareri				x

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Family Idoteidae				
Idotea wosnesenskii			x	x
Family Asellidae				
Ianiropsis kincaidi kincaidi				x
Family Munnidae				
Munna sp.				x
Munna stephenseni				x
Order Amphipoda		x	x	x
Suborder Gammaridea				
Family Ampithoidae				
Amphithoe sp.			x	x
Ampithoe similans		x	x	x
Ampithoe rubricatoides		x		
Family Calliopiidae				
Oligochinus lighti		x	x	x
Calliopiella pratti			x	
Family Corophiidae				
Corophium sp.		x		
Family Eusiridae				
Accedomoera vagor		x		
Accedomoera sp.		x		
Paramoera columbiana			x	x
Pontogeneia sp.			x	
Family Gammaridae				
Anisogammarus locustoides	x			
Anisogammarus sp.		x		
Melita sp.			x	

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island									
Family Hyalellidae													
Najna conciliorum				x									
Family Hyalidae													
Allorchestes angustus		x											
Hyale rubra				x									
Hyale rubra frequens				x									
Parallorchestes ochotensis			x	x									
Family Ischyroceridae													
Ischyrocerus sp.		x											
Ischyrocerus anguipes		x	x										
Jassa pulcella		x	x										
Family Lysianassidae													
Anonyx sp.				x									
Family Pleustidae													
Parapleustes nautilus				x									
Parapleustes sp.			x										
Stenopleustes uncigera		x											
Family Stenothoidae		x											
Metopa sp.			x										
Suborder Caprellidea													
Family Caprellidae	x	x	x	x									
Superorder Eucarida													
Order Decapoda													
Pagurus sp.				x									
Pagurus beringanus			x										
Pagurus h. hirsutiusculus		x	x	x									
Pugettia gracilis			x										
Telmessus cheiragonus		x											
Cancer sp.			x										

Appendix Table IIA.--(continued).

	Cape Peirce	Crooked Island	Nukshak Island	Spectacle Island
Class Insecta			x	
Order Collembola				
Anurida maritima				x
Order Coleoptera	x	x	x	x
Family Staphylinidae				x
Order Diptera	x		x	x
Family Chironomidae	x		x	x
PHYLUM SIPUNCULA			x	
PHYLUM ECHURA				
Bonelliopsis alaskana				x
PHYLUM ECTOPROCTA	x		x	
Microporina borealis				x
PHYLUM ECHINODERMATA				
Class Asteroidea				
Leptasterias hexactis			x	x
Leptasterias sp.			x	
Class Ophiuroidea			x	x
Class Echinoidea				
Strongylocentrotus droebachiensis			x	
Class Holothuroidea				
Cucumaria pseudocurata			x	x
PHYLUM UROCHORDATA				x

Appendix Table IIB.--List of the species of benthic plants and invertebrates at intertidal sites in Bristol Bay and the Aleutian Islands

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
PHYLUM CYANOPHYTA						X							
PHYLUM CHLOROPHYTA			X	X									
Family Prasiolaceae													
Rosenvingiella polyrhiza										X			
Family Monostromataceae													
Monostroma sp.							X	X			X	X	X
Monostroma fuscum							X			X	X	X	X
Monostroma zostericola											X		
Family Ulvaceae													
Ulva sp.													X
Ulva lactuca											X		X
Enteromorpha sp.												X	
Enteromorpha intestinalis		X											
Family Acrosiphoniaceae													
Spongomorpha sp.												X	
Spongomorpha cf. *mertensii*												X	
Spongomorpha spinescens							X			X	X	X	X
Family Cladophoraceae													
Chaetomorpha sp.											X		
Cladophora sp.					X						X	X	
Cladophora flexuosa									X				X
Cladophora seriacea													X
Lola lubrica						X				X			
Rhizoclonium riparium													X
PHYLUM CRYSOPHYTA													
Subphylum Bacillariophyceae		X								X		X	

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
PHYLUM PHAEOPHYTA	x												
Order Ectocarpales													
Family Ectocarpaceae				x		x							
Pylaiella littoralis											x		
Ectocarpus simulans													x
Ectocarpus sp.												x	
Family Ralfsiaceae													
Ralfsia fungiformes							x				x		
Family Elachistaceae						x							
Family Corynophlaeaceae													
Leathesia difformis						x							
Family Chordariaceae						x							
Analipus japonicus							x					x	
Order Sphacelariales													
Family Sphacelariaceae													
Sphacelaria sp.													x
Order Laminariales													
Family Laminariaceae													
Laminaria sp.							x				x		
Laminaria longipes							x				x		
Laminaria groenlandica							x						
Hedophyllum sessile											x		
Family Alariaceae													
Alaria sp.							x			x	x	x	x
Alaria marginata											x		x
Alaria taeniata							x				x		x
Alaria praelonga									x				
Alaria cf. *crispa*												x	

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Alariaceae - cont'd.													
Desmarestia sp.								x					x
Desmarestia viridis													x
Family Fucaceae													
Fucus sp.						x						x	
Fucus distichus							x	x			x	x	x
Order Scytosiphonales													
Family Scytosiphonaceae													
Colpomenia bullosa												x	
Scytosiphon lomentaria							x			x			
PHYLUM RHODOPHYTA													
Family Bangiaceae													
Bangia fuscopurpurea								x	x				
Porphyra sp.		x					x	x	x	x	x	x	x
Family Cruoriaceae													
Petrocelis franciscana											x		
Petrocelis sp.													x
Family Phyllophoraceae													
Ahnfeltia sp.					x	x							
Ahnfeltia plicata					x								
Family Gigartinaceae													
Iridaea sp.							x					x	
Iridaea cornucopiae							x				x		x
Gigartina sp.												x	
Order Cryptonemiales													
Family Dumontiaceae													
Cryptosiphonia woodii		x									x		

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Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Endocladiaceae													
Endocladia muricata											x	x	x
Gloiopeltis furcata													x
Family Corallinaceae				x								x	
Corallina sp.							x				x		
Corallina vancouverensis										x			
Bossiella sp.							x		x	x	x		
Bossiella chiloensis							x				x		
Bossiella plumosa											x		
Family Kallymeniaceae													
Callophyllis sp.									x				x
Callophyllis flabellulata									x				
Family Rhodymeniaceae													
Rhodymenia sp.										x	x	x	
Rhodymenia palmata										x		x	
Palmaria palmata											x	x	x
Halosaccion glandiforme							x		x		x		x
Halosaccion sp.										x		x	
Family Ceramiaceae													
Antithamnion sp.												x	
Callithamnion pikeanum									x		x		
Microcladia borealis											x		
Microcladia coulteri											x		
Ptilota sp.							x				x		
Ptilota felicinia										x			
Neoptilota sp.							x		x				
Neoptilota asplenoides							x						
Family Delesseriaceae													
Tokidadendron bullata							x					x	
Hypophyllum sessile											x		
Family Rhodomelaceae			x										
Pterosiphonia sp.						x	x					x	
Pterosiphonia bipinnata				x			x	x	x	x	x	x	x

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Rhodomelaceae - cont'd.													
Rhodomela larix										x			
Odonthalia sp.										x	x		x
Odonthalia floccosa				x			x		x		x	x	x
Odonthalia washingtoniensis							x	x					
PHYLUM ANTHOPHYTA													
Class Monocotyledoneae													
Zostera marina	x		x	x	x	x							
PHYLUM PROTOZOA													
Class Foraminifera	x												
PHYLUM PORIFERA					x	x	x			x	x	x	x
PHYLUM CNIDARIA													
Class Hydrozoa						x							
Order Hydroidea		x											
Family Sertulariidae													
Obelia sp.												x	
Sertularia sp.								x				x	
Abietinaria filicula								x					
Sertularia cupressoides												x	
Family Haleciidae													
Halecium sp.					x								
Class Anthozoa		x		x	x	x	x	x	x			x	x
Order Alcyonacea													
Eunephyta rubiformis											x		

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
PHYLUM PLATYHELMINTHES								x		x		x	
Class Turbellaria							x		x		x		x
PHYLUM RHYNCHOCOELA (NEMERTEA)	x	x	x	x	x	x	x		x	x	x	x	
Emplectonema gracile								x					x
PHYLUM NEMATODA	x	x	x	x	x	x	x	x	x	x	x	x	x
PHYLUM ANNELIDA													
Class Polychaeta													
Family Polynoidae								x	x				x
Harmothoe sp.		x											
Harmothoe imbricata	x			x	x	x					x		
Hesperone sp.					x								
Family Sigalionidae													
Pholoe minuta	x	x		x	x	x		x			x		x
Family Chrysopetalidae													
Paleonotus bellis												x	
Dysponetus sp.											x	x	
Dysponetus pygmaeus												x	
Family Phyllodocidae													
Anaitides groenlandica				x	x	x							
Anaitides maculata	x	x		x	x	x							
Anaitides sp.				x							x		
Eteone longa	x	x		x	x	x		x		x	x	x	x
Eteone pacifica	x	x			x		x				x		x
Eulalia sp.					x		x				x		x
Eulalia quadrioculata						x	x		x	x	x	x	x
Eulalia bilineata				x		x	x						
Mystides borealis										x			

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Syllidae													
Autolytus sp.				x	x						x	x	x
Typosyllis sp.					x	x			x		x	x	
Typosyllis variegata										x		x	
Typosyllis alternata							x			x	x	x	x
Typosyllis pulchra							x		x	x	x	x	x
Typosyllis elongata					x							x	
Typosyllis a. adamantea								x			x	x	x
Exogone gemmifera	x			x	x	x	x					x	
Exogone sp.				x	x	x					x	x	
Exogone verugera					x								
Sphaerosyllis sp.	x			x	x					x	x	x	x
Sphaerosyllis pirifera	x	x		x	x	x							x
Subfamily Parasphaerosyllis												x	
Family Nereidae													
Nereis sp.					x	x	x			x	x	x	x
Nereis pelagica							x		x		x	x	
Nereis vexillosa								x				x	
Nereis zonata					x						x		
Family Nephtyidae													
Nephtys sp.	x	x		x		x							
Nephtys caeca	x	x		x	x	x							
Nephtys ciliata				x	x								
Family Sphaerodoridae													
Sphaerodoropsis minuta				x	x	x	x				x	x	x
Sphaerodoropsis sphaerulifer								x					
Family Glyceridae	x												
Glycera capitata											x		x
Family Goniadidae													
Glycinde picta	x	x											x
Goniada annulata				x									

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Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Lumbrineridae													
Lumbrineris sp.						x	x				x		
Lumbrineris inflata										x	x	x	
Family Orbiniidae										x			
Haploscoloplos elongatus	x	x				x							
Haploscoloplos sp.						x							
Haploscoloplos panamensis	x			x	x	x							
Naineris quadricuspida						x					x	x	
Naineris laevigata							x				x		
Naineris sp.						x	x						x
Scoloplos armiger				x									
Family Paraonidae													
Aricidea suecica	x	x											
Aricidea sp.				x	x	x							
Family Spionidae										x			
Laonice cirrata			x										
Nerine cirratulus											x		
Polydora sp.	x	x		x								x	
Polydora socialis	x	x											
Polydora caulleryi					x								
Polydora ciliata				x							x		
Polydora quadrilobata				x	x	x		x				x	
Spio filicornis	x	x		x	x						x		
Boccardia natrix											x		x
Spiophanes bombyx	x	x											
Rhyncospio sp.	x	x	x	x	x	x						x	
Pygospio elegans	x												
Family Cirratulidae													
Cirratulus cirratus	x	x				x	x		x	x	x	x	x
Cirratulus sp.											x		
Caulleriella sp.		x											
Tharyx multifilis	x	x											
Tharyx sp.	x			x	x	x						x	x
Chaetozone setosa	x	x		x	x	x							

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Flabelligeridae													
Brada villosa					x								
Flabelligera affinis												x	
Family Scalibregmidae													
Scalibregma inflatum											x	x	
Family Capitellidae													
Capitella capitata	x	x		x	x	x	x		x	x	x	x	x
Heteromastus filiformis	x	x		x									x
Heteromastus sp.													x
Family Arenicolidae					x	x							
Abarenicola pacifica	x	x											
Arenicola gracilis	x	x											
Family Maldanidae	x		x									x	
Praxillella affinis		x		x	x	x							
Family Oweniidae													
Owenia fusiformis	x	x		x	x	x							
Family Pectinariidae													
Cistenides brevicoma		x				x							
Cistenides sp.					x								
Cistenides granulata				x									
Family Ampharetidae							x						
Ampharete arctica	x	x		x	x	x							
Asabellides sibirica					x		x				x	x	x
Pseudosabellides sp.							x						
Family Terebellidae													
Nicolea zostericola												x	
Polycirrus medusa	x	x					x				x	x	x
Family Trichobranchidae													
Terebellides stroemii				x	x	x							

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Sabellidae		x											
Chone gracilis				x	x		x		x		x	x	x
Chone sp.				x								x	
Chone cincta				x	x	x						x	
Pseudopotamilla reniformis				x		x						x	
Potamilla neglecta												x	
Schizobranchia insignis									x		x	x	
Amphiglena sp.												x	
Amphiglena pacifica				x			x				x		x
Fabricia sabella				x	x	x	x	x		x	x	x	x
Fabricia sp.										x			x
Fabricia pacifica													x
Fabricia crenicolis							x	x		x	x	x	x
Laonome sp.				x	x	x							
Family Serpulidae					x								
Dexiospira spirillum										x	x		
Dexiospira sp.									x				
Family Saccocirridae													
Saccocirrus sp.						x							
Class Oligochaeta			x	x		x		x		x	x	x	x
Family Enchytraeidae	x	x					x		x		x		x
PHYLUM MOLLUSCA													
Class Polyplacophora							x			x	x	x	x
Cyanoplax dentiens							x				x		x
Ischnochiton abyssicola													x
Tonicella lineata							x			x		x	
Spongioradsia aleutica												x	
Schizoplax brandtii							x				x		
Katharina tunicata							x		x	x	x	x	x
Mopalia ciliata												x	x
Class Aplacophora													
Chaetoderma robusta					x								

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Class Bivalvia													
Mytilus edulis	x	x		x				x	x	x	x	x	x
Musculus discors							x		x		x	x	
Musculus vernicosus							x				x		
Musculus olivaceous												x	
Musculus sp.							x					x	x
Modiolus modiolus							x				x	x	x
Axinopsida serricata	x	x			x	x							
Kellia laperousi											x		
Kellia sp.											x		
Mysella tumida											x	x	
Turtonia occidentalis					x	x	x	x	x		x	x	x
Clinocardium nuttallii		x											
Macoma sp.					x	x					x	x	
Macoma brota					x	x							
Macoma obliqua		x											
Macoma balthica	x	x		x									
Protothaca staminea											x		x
Mya sp.	x												
Mya arenaria	x	x											
Hiatella arctica	x				x		x		x	x	x	x	x
Class Gastropoda													
Acmaea sybaritica							x						
Collisella sp.		x								x	x	x	
Collisella pelta							x	x	x	x	x	x	x
Collisella cf. *strigatella*										x			
Collisella digitalis									x	x			x
Notoacmea scutum							x				x	x	
Notoacmea persona											x		
Margarites sp.				x	x	x				x	x		
Margarites helicinus							x				x		x
Margarites cf. *helicinus*										x			
Margarites beringensis											x	x	
Moellaria sp.											x		
Moellaria drusiana													x
Moellaria quadrai											x		
Halochoncha reflexa					x		x				x		
Lacuna variegata										x			

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Class Gastropoda													
Lacuna marmorata									x	x	x		x
Lacuna vincta	x						x			x			x
Littorina sp.					x							x	
Littorina scutulata					x	x							x
Littorina aleutica							x		x				
Littorina sitkana		x				x	x	x	x	x	x	x	x
Alvinia aurivillii											x		x
Cingula sp.													x
Cingula aleutica												x	x
Barleeia subtenuis													x
Barleeia sp.							x	x	x		x		x
Cerithiopsis sp.											x		
Melanella sp.											x		
Melanella randolfi											x	x	x
Balcis sp.										x			
Balcis alaskensis												x	
Trichotropis insignis							x						
Nucella sp.											x		
Nucella canaliculata											x	x	x
Nucella lamellosa							x						x
Nucella lima							x	x		x	x	x	x
Mitrella rosacea											x		
Mitrella cf. *rosacea*												x	
Buccinum baeri							x				x	x	x
Buccinum sp.												x	
Odostomia sp.		x					x				x		x
Cylichna sp.				x									
Siphonaria thersites											x		x
Order Nudibranchia						x							
Onchidella borealis													x
PHYLUM ARTHROPODA													
Class Pseudoscorpionida													x

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Class Acarina							x					x	x
Family Halicaridae											x		x
Class Pycnogonida											x		
Nymphon grossipes					x								
Ammothea alaskensis												x	x
Ammothea gracilipes													x
Ammothea latifrons				x	x	x						x	
Ammothea pribilofensis							x			x		x	x
Achelia chelata							x					x	
Achelia spinosa													x
Phoxichilidium femoratum					x	x							
Pycnogonum sp.													x
Class Crustacea				x									
Subclass Platycopa	x												x
Subclass Copepoda												x	
Order Harpacticoida	x	x				x							
Family Peltidiidae													x
Subclass Cirripedia													
Order Thoracica													
Chthamalus dalli							x		x	x	x	x	x
Balanus sp.							x	x	x			x	
Balanus balanus											x		
Balanus cariosus							x		x	x	x	x	x
Balanus glandula							x	x	x	x	x	x	x
Subclass Malacostraca													
Order Mysidacea													

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Mysidae					x								
Order Cumacea				x	x	x		x					
Campylaspis affinis		x									x		
Family Nannastacidae						x							
Cumella sp.	x										x		
Order Tanaidacea	x			x					x				
Order Isopoda													
Family Sphaeromatidae										x			
Gnorimosphaeroma oregonensis													x
Exosphaeroma amplicauda												x	x
Dynamenella sheareri												x	x
Family Idoteidae													
Saduria entomon		x	x										
Idotea fewkesi							x			x		x	
Edotea sp.			x										
Idotea wosnesenskii							x	x	x		x	x	x
Idotea sp.							x						
Family Asellidae													
Ianiropsis kincaidi kincaidi		x								x			
Family Munnidae													
Munna stephenseni									x		x	x	
Munna chromatocephala										x		x	
Family Dajidae													
Cryptothir balani											x		

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Order Amphipoda													
Suborder Gammaridea													
Family Ampithoidae				x									x
Ampithoe sp.	x			x							x		x
Ampithoe similans							x			x	x	x	x
Ampithoe rubricatoides				x									
Family Calliopiidae													
Oligochinus lighti											x	x	x
Calliopiella pratti									x		x		x
Family Corophiidae													
Corophium sp.	x			x	x	x				x			
Family Eusiridae													
Accedomoera vagor							x			x	x		
Paramoera columbiana								x		x		x	
Pontogeneia sp.							x						
Family Gammaridae													
Anisogammarus pugettensis		x											
Anisogammarus locustoides		x											
Gammarus locusta	x												
Melita sp.											x		
Anisogammarus sp.						x							
Family Haustoriidae													
Pontoporeia affinis			x										
Family Hyalellidae													
Najna conciliorum							x						
Family Hyalidae								x					x
Allorchestes angustus										x			
Hyale rubra											x		
Parallorchestes ochotensis							x		x	x	x	x	

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Family Isaeidae													
Protomedeia grandimana	x												
Family Ischyroceridae													
Ischyrocerus sp.							x				x		
Ischyrocerus anguipes							x	x					x
Ischyrocerus cf. *tsvetkovae*												x	
Jassa pulcella													
Family Lysianassidae			x										
Anonyx sp.									x		x		
Orchomene sp.													x
Family Phoxocephalidae													
Paraphoxus sp.											x		
Family Pleustidae													
Parapleustes nautilus										x	x	x	x
Parapleustes sp.												x	
Family Stenothoidae												x	
Suborder Caprellidea													
Family Caprellidae	x	x		x	x	x	x			x	x	x	
Superorder Eucarida													
Order Decapoda						x							
Crangon sp.		x			x								
Pagurus sp.	x											x	x
Pagurus beringanus										x		x	
Pagurus h. hirsutiusculus							x			x	x		x
Haplogaster grebnitzkii											x		
Telmessus cheiragonus				x									
Cancer oregonensis											x	x	

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Class Insecta													
Order Collembola													
Anurida maritima													x
Order Coleoptera							x				x		x
Family Staphylinidae									x		x	x	x
Order Diptera								x	x	x	x	x	x
Suborder Cyclorrapha												x	
Family Chironomidae		x		x					x		x	x	x
PHYLUM SIPUNCULA													
Family Sipunculidae											x		
PHYLUM ECHIURA													
Bonelliopsis alaskana											x		
PHYLUM PRIAPULIDA													
Priapulus caudatus				x	x	x							
PHYLUM ECTOPROCTA				x		x							
Flustrella sp.											x		
Dendrobeania murryana												x	
Scrupocellaria arctica												x	
Notoplites sp.												x	
PHYLUM BRACHIPODA													x
PHYLUM ECHINODERMATA													
Class Asteroidea						x							
Leptasterias alaskensis												x	
Leptasterias camtschatica												x	
Leptasterias hexactis							x		x		x	x	x

Appendix Table IIB.--(continued).

	Middle Point	Point Edward	Moffett Lagoon	Izembek Lagoon	Blaine Point	Cape Glazenap	Amak Island	Cape Lapin	Cape Mordvinof	Sennet Point	Akun Island	Eider Point	Portage Bay
Class Asteroidea - cont'd.													
Leptasterias leptodoma												x	
Leptasterias sp.												x	
Henricia laeviuscula											x	x	
Class Ophiuroidea					x		x						
Ophiopholis aculeata							x						
Class Echinoidea													
Strongylocentrotus sp.											x	x	
Strongylocentrotus droebachiensis							x					x	
Class Holothuroidea				x	x	x							
Cucumaria pseudocurata							x	x	x	x	x	x	x
Pseudostichopus aleutians													x
Family Phyllophoridae													x
PHYLUM UROCHORDATA													
Class Ascidiacea				x									
Styela clavata											x		
Molgula oregonia					x	x						x	

Appendix Table IIC.--Presence (x) or absence of benthic plants and invertebrates at intertidal sites in the Pribilof Islands.

	Otter Island	English Bay, St.Paul Is.	High Bluffs, St.George Is	Garden Cove, St.George Is	Zapadni Bay, St.George Is
PHYLUM CHLOROPHYTA					
Family Prasiolaceae					
Prasiola sp.			x		
Rosenvingiella polyrhiza			x		
Family Monostromataceae					
Monostroma sp.			x		
Monostroma arcticum			x		
Monostroma fuscum	x	x			x
Family Ulvaceae					
Enteromorpha sp.			x	x	
Family Acrosiphoniaceae					
Spongomorpha sp.	x			x	
Spongomorpha arcta	x			x	
Spongomorpha mertensii			x		
Spongomorpha cf. *S. saxatilis*	x		x		
Spongomorpha spinescens			x		
Urospora sp.	x		x	x	
Urospora mirabilis			x	x	
Urospora wormskioldii	x		x		
Family Cladophoraceae					
Cladophora sp.	x			x	x
Cladophora gracilis	x				
Cladophora seriacea					x
Rhizoclonium riparium			x		
Lola lubrica				x	
Lola/Rhizoclonium sp.	x				
Rhizoclonium implexum	x				
PHYLUM CRYSOPHYTA					
Subphylum Bacillariophyceae			x		
Licmophora sp.	x			x	

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Appendix Table IIC.--(continued).

	Otter Island	English Bay, St. Paul Is.	High Bluffs, St. George Is	Garden Cove, St. George Is	Zapadni Bay, St. George Is
PHYLUM PHAEOPHYTA					
Family Ectocarpaceae					
Pylaiella littoralis	x		x		x
Family Ralfsiaceae					
Ralfsia sp.	x				
Ralfsia fungiformis					x
Family Chordariaceae					
Analipus filiformis		x			
Family Punctariaceae					
Melanosiphon sp.			x		
Family Scytosiphonaceae					
Petalonia fascia					x
Scytosiphon lomentaria	x				
Family Laminariaceae					
Laminaria sp.			x		x
Laminaria groenlandica	x	x			x
Laminaria longipes		x			
Family Alariaceae					
Alaria sp.	x	x	x		x
Alaria fistulosa	x				
Alaria taeniata	x				x
Family Fucaceae					
Fucus sp.			x		
Fucus distichus	x		x	x	x
PHYLUM RHODOPHYTA					
Family Bangiaceae					
Porphyra sp.	x		x		x

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is								
Family Gigartinaceae													
Iridaea sp.	x	x	x	x	x								
Iridaea cornucopiae	x	x			x								
Family Endocladiaceae													
Gloiopeltis furcata	x												
Family Corallinaceae													
Bossiella cretacea		x											
Corallina sp.	x												
Corallina vancouveriensis		x											
Lithothamnium sp.	x				x								
Subfamily Melobesioideae		x											
Family Kallymeniaceae													
Callophyllis crustata		x											
Callophyllis flabellulata	x												
Family Choreocolacaceae													
Harveyella? sp.		x											
Family Rhodymeniaceae													
Halosaccion sp.			x	x									
Halosaccion glandiforme	x		x	x	x								
Rhodymenia sp.	x		x	x	x								
Palmaria palmata	x				x								
Family Ceramiaceae													
Ptilota sp.				x	x								
Ptilota filicina		x	x		x								
Neoptilota sp.		x											
Family Delesseriaceae			x										
Nienburgia sp.					x								
Family Rhodomelaceae													
Polysiphonia brodiaei		x											
Pterosiphonia bipinnata	x				x								

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is								
Family Rhodomelaceae - cont'd.													
Rhodomela larix		x											
Odonthalia sp.		x			x								
Odonthalia floccosa		x											
Odonthalia kamschatica		x											
PHYLUM ANTHOPHYTA													
Family Potamogetonaceae		x											
PHYLUM PORIFERA			x		x								
Class Demospongiae													
Halichondria sp.	x	x											
Halichondria panicea		x											
PHYLUM CNIDARIA													
Class Hydrozoa													
Family Haleciidae													
Halecium sp.					x								
Family Eudendriidae													
Eudendrium sp.					x								
Eudendrium annulatum					x								
Family Tubulariidae													
Tubularia simplex	x												
Family Campanulinidae					x								
Family Sertulariidae													
Sertularia sp.					x								
Sertularia albimaris					x								
Abietinaria sp.					x								
Class Anthozoa					x								
Order Actiniaria	x	x											

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
PHYLUM PLATYHELMINTHES					
Class Turbellaria					x
Order Polycladida		x			
PHYLUM RHYNCHOCOELA (NEMERTEA)	x	x			x
PHYLUM NEMATODA	x	x	x		x
PHYLUM ANNELIDA					
Class Polychaeta					
Family Polynoidae					
Arctonoe vittata		x			
Harmothoe imbricata		x			x
Family Pholoididae					
*Pholoides** (=*Peisidice*)*aspera*		x			
Family Chrysopetalidae					
Dysponetus sp.		x			
Family Phyllodocidae					
Anaitides mucosa					x
Eteone sp.					x
Eteone longa	x	x			x
Eulalia bilineata		x			
Family Syllidae					
Autolytus sp.	x				x
Typosyllis sp.	x	x			x
Typosyllis alternata					x
Typosyllis pulchra					x
Typosyllis variegata	x	x			
Typosyllis hyalina		x			

* Fauchald (1977)

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St. Paul Is.	High Bluffs, St. George Is	Garden Cove, St. George Is	Zapadni Bay, St. George Is
Family Syllidae - cont'd.					
Eusyllis sp.		x			
Eusyllis assimilis					x
Exogone gemmifera	x		x		x
Exogone verugera		x			
Sphaerosyllis sp.	x	x			
Sphaerosyllis pirifera	x	x			x
Parasphaerosyllis sp.					x
Syllides japonica		x			x
Family Nereidae					
Nereis sp.	x	x			x
Nereis pelagica	x	x			x
Nereis vexillosa					x
Family Orbiniidae					
Naineris sp.					x
Naineris quadricuspida		x			x
Naineris laevigata					x
Family Paraonidae					
Aricidea sp.				x	
Family Spionidae					
Polydora sp.		x			x
Polydora quadrilobata	x				
Polydora quadricuspida					x
Spio filicornis		x			x
Boccardia sp.		x			
Boccardia natrix		x			x
Boccardia proboscidea	x				x
Family Cirratulidae					
Cirratulus cirratus	x	x	x		x
Cirratulus sp.					x
Tharyx sp.		x			x
Family Capitellidae					
Capitella capitata	x				x

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
Family Maldanidae					x
Family Oweniidae					
Owenia fusiformis		x			
Family Ampharetidae					
Asabellides sibirica	x	x			x
Family Terebellidae					
Nicolea zostericola					x
Polycirrus medusa	x	x			
Family Sabellidae					
Chone gracilis	x				x
Chone cincta	x	x			
Chone sp.					x
Potamilla neglecta	x				x
Amphiglena sp.	x	x			
Amphiglena pacifica	x				x
Fabricia sabella	x	x		x	x
Oriopsis minuta *	x	x			x
Fabriciola berkeleyi †	x				
Fabricia crenicollis	x	x			x
Class Oligochaeta	x	x	x	x	
Family Enchytraeidae	x				x
PHYLUM MOLLUSCA					
Class Polyplacophora					
Tonicella rubra	x				
Katharina tunicata					x
Schizoplax brandtii	x	x			x
Class Bivalvia					
Nuculana pernula					x
Mytilus edulis	x				x
Musculus sp.					x

* *O. minuta* = *Fabricia minuta*, auctt. (Hobson and Banse, in press).

† *F. berkeleyi* = *Fabricia pacifica*, auctt. (Hobson and Banse, in press).

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
Class Bivalvia - cont'd.					
Musculus discors	x				x
Musculus vernicosus					x
Modiolus modiolus	x	x			
Mysella sp.		x			
Mysella tumida		x			
Turtonia sp.		x			
Turtonia occidentalis	x	x			x
Saxidomus gigantea		x			
Protothaca staminea					x
Hiatella arctica	x	x			x
Class Gastropoda					
Acmaea mitra		x			
Collisella pelta		x			x
Collisella cf. *C. strigatella*		x			
Cryptobranchia concentrica		x			
Margarites sp.		x			
Margarites helicinus	x	x			x
Margarites beringensis		x			x
Littorina sitkana	x	x	x	x	x
Littorina sp.	x				x
Haloconcha reflexa	x	x	x		x
Lacuna variegata	x				x
Alvinia sp.		x			
Cingula aleutica		x			
Cingula kyskensis	x				
Cingula sp.	x	x			
Cingula katharina		x			
Barleeia sp.	x				
Nucella sp.		x			
Nucella canaliculata		x			
Nucella lima	x	x			x
Buccinum baeri	x				x
Volutharpa ampullacea	x				
Mitrella gouldi					x
Mitrella rosacea					x

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
PHYLUM ARTHROPODA					
Class Acarina			x	x	
Family Halacaridae	x				x
Class Pycnogonida					
Ammothea latifrons		x			
Ammothea pribilofensis		x			x
Achelia chelata					x
Tanystylum anthomasti	x				
Class Crustacea					
Subclass Copepoda					x
Nebalia sp.					x
Order Harpacticoida	x	x	x	x	x
Subclass Cirripedia					
Balanus sp.					x
Balanus cariosus					x
Balanus glandula	x				
Subclass Malacostraca					
Order Mysidacea					x
Mysis sp.	x				
Archeomysis grebnitzkii			x		
Order Tanaidacea					
Family Tanaidae	x				
Order Isopoda					
Idotea wosnesenskii	x				x
Idotea fewkesi	x	x			x
Munna sp.	x				x
Munna chromatocephala	x	x			

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
Order Amphipoda					x
Suborder Gammaridea					
Family Acanthonotozomatidae					
Odius carinatus					x
Family Ampithoidae					
Ampithoe sp.		x			x
Ampithoe rubricatoides					x
Family Calliopiidae					
Oligochinus lighti		x			
Calliopiella pratti	x				x
Family Eusiridae					
Accedomoera vagor					x
Paramoera columbiana		x		x	
Pontogeneia sp.					x
Family Hyalidae					
Allorchestes maleolus					x
Hyale rubra	x				
Parallorchestes ochotensis	x	x	x		x
Family Ischyroceridae					
Ischyrocerus sp.	x				x
Ischyrocerus anguipes	x				x
Ischyrocerus tsvetkovae					x
Jassa pulcella	x	x			
Family Lysianassidae					
Anonyx sp.		x			x
Orchomene sp.		x			x
Family Phoxocephalidae					x

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
Family Pleustidae					
Parapleustes johanseni					x
Sympleustes suberitobius					x
Stenopleustes uncigera					x
Family Stenothoidae	x	x			x
Suborder Caprellidea					
Family Caprellidae		x			
Caprella sp.					x
Caprella cristibrachium					x
Class Insecta					
Order Diptera			x	x	
Order Coleoptera	x				
Family Staphylinidae					x
PHYLUM SIPUNCULIDA		x			
PHYLUM BRYOZOA					
Hippothoa hyalina		x			x
Flustrella gigantea		x			
Dendrobeania murrayana	x				
PHYLUM ECHINODERMATA					
Class Asteroidea					
Leptasterias hexactis		x			
Pisaster ochraceus					x
Class Echinoidea					
Strongylocentrotus droebachiensis					x
Class Ophiuroidea					x

Appendix Table IIC.--(continued).

	Otter Island	English Bay, St.Paul Is.	High Bluffs,St.George Is	Garden Cove,St.George Is	Zapadni Bay,St.George Is
Class Holothuroidea					
Cucumaria pseudocurata	x	x	x		x
PHYLUM UROCHORDATA					
Class Ascidiacea					
Molgula oregonia	x				x
Sigillinaria sp.					x

Appendix Table IID.--List of the species of benthic plants and invertebrates at intertidal and subtidal sites in the Norton Sound Region.

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
PHYLUM CYANOPHYTA		x										x	
Family Oscillatoriaceae			x		x				x				
Oscillatoriaceae cf. *Lyngbya*										x			
Family Rivulariaceae	x										x		
PHYLUM CHLOROPHYTA							x	x	x	x			
Family Monostromataceae													
Monostroma sp.	x												
Family Ulvaceae													
Ulva lactuca	x												
Enteromorpha sp.		x	x	x									
Enteromorpha intestinalis	x												
Enteromorpha linza	x												
Enteromorpha clathrata	x	x											
Family Acrosiphoniaceae													
Spongomorpha cf. *S. saxatilis*	x												
Family Cladophoraceae													
Lola lubrica	x												
PHYLUM CRYSOPHYTA													
Subphylum Bacillariophyceae	x	x				x	x		x				
PHYLUM PHAEOPHYTA													
Order Ectocarpales													
Family Ectocarpaceae													
Pylaiella littoralis	x	x											
Ectocarpus sp.										x			

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Order Dictyosiphonales								x					
Family Punctariaceae													
Melanosiphon intestinalis	x	x	x										
Order Sphacelariales													
Family Sphacelariaceae													
Sphacelaria racemosa		x								x			
Order Laminariales													
Family Laminariaceae													
Laminaria sp. §													x
Agarum sp.													x
Family Alariaceae													
Alaria sp. §													x
Order Fucales													
Family Fucaceae													
Fucus sp.		x		x		x				x	x	x	
Fucus distichus	x	x											
Order Scytosiphonales													
Family Scytosiphonaceae													
Petalonia fascia	x												
Scytosiphon sp.	x												
PHYLUM RHODOPHYTA											x		
Family Bangiaceae													
Porphyra sp.	x												
Family Acrochaetiaceae													
Acrochaetium sp.	x	x											

§. Collected from subtidal region.

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Family Phyllophoraceae													
Ahnfeltia plicata		x								x			
Family Gigartinaceae								x					
Iridaea sp.		x											
Gigartina pacifica	x												
Family Rhodymeniaceae		x											
Halosaccion glandiforme													x
Family Ceramiaceae													
Ptilota sp.		x											
Family Rhodomelaceae					x				x	x			
Pterosiphonia sp.		x											
Pterosiphonia bipinnata	x	x											
PHYLUM ANTHOPHYTA		x											
Family Potamogetonaceae		x		x	x	x			x	x		x	
PHYLUM PORIFERA				x									
Class Demospongiae													
Halichondria sp.										x			
PHYLUM CNIDARIA													
Class Hydrozoa													
Family Sertulariidae													
Obelia longissima										x			
Sertularia albimaris													
Abietinaria sp.		x											
Abietinaria filicula										x			
Thuiaria sp.		x											
Family Halciidae													
Halecium sp.		x											

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Class Scyphozoa													
Haliclystus sp.								x					
Class Anthozoa													
Order Alcyonacea		x											
Eunephtya rubiformis ¶										x			
PHYLUM RHYNCHOCOELA (NEMERTEA)			x										
PHYLUM NEMATODA	x	x				x							
PHYLUM ANNELIDA													
Class Polychaeta													
Family Phyllodocidae													
Eteone longa	x		x	x			x	x			x		
Family Goniadidae													
Glycinde picta			x						x				
Family Orbiniidae													
Naineris quadricuspida						x							
Family Spionidae													
Spio filicornis	x												
Rhyncospio sp.			x										
Pygospio sp.			x				x		x				
Pygospio elegans			x										
Family Cirratulidae		x											
Cirratulus cirratus						x						x	
Tharyx sp.						x							
Family Capitellidae	x												
Capitella capitata		x										x	

¶. Probably washed ashore from the sublittoral region.

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Family Arenicolidae													
Arenicola glacialis			X						X				
Family Ampharetidae													
Asabellides sibirica								X					
Family Sabellidae													
Amphiglena sp.							X						
Fabricia sabella	X												
Family Saccocirridae													
Saccocirrus sp.											X		
Class Oligochaeta	X	X	X		X	X	X	X			X		
PHYLUM MOLLUSCA													
Class Bivalvia									X				
Mytilus edulis	X					X		X		X			
Musculus sp.		X											
Turtonia occidentalis			X										
Class Gastropoda				X									
Littorina sitkana	X									X			
Littorina squalida	X												
Littorina sp.											X		
Lacuna variegata	X												
Lacuna vincta	X												
Lacuna sp.	X												
PHYLUM ARTHROPODA													
Class Acarina	X												
Class Crustacea													
Subclass Copepoda			X										

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Subclass Cirripedia											x		
Chthamalus dalli	x	x											
Family Balanidae	x												
Subclass Malacostraca													
Order Mysidacea													
Neomysis czerniawskii				x									
Neomysis sp.					x								
Order Cumacea							x						
Order Isopoda												x	
Saduria entomon		x	x										
Order Amphipoda	x	x					x						
Suborder Gammaridea													
Family Eusiridae													
Paramoera columbiana							x						
Family Gammaridae													
Anisogammarus sp.	x	x					x			x			
Gammarus sp.	x				x							x	
Family Haustoriidae													
Pontoporeia affinis				x					x				
Family Lysianassidae													
Orchomene sp.				x									
Onisimus sp.				x									
Onisimus littoralis				x						x			
Suborder Caprellidea													
Family Caprellidae	x												

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
Superorder Eucarida													
Order Decapoda													
Crangon sp.			X	X									
Paralithodes platypus §													X
Hyas sp. §													X
Class Insecta													
Order Collembola													
Anurida maritima											X		
Order Hemiptera						X		X					
Suborder Homoptera								X					
Family Psyllidae							X						
Order Hymenoptera							X						
Order Coleoptera		X											
Family Staphylinidae													
Order Diptera	X	X									X		
Family Chironomidae	X	X	X		X								
Family Culicidae	X										X	X	
PHYLUM ECTOPROCTA							X					X	
Alcyonidium disciforme										X			
Flustrella gigantea										X			
Eucratea loricata		X											

§. Collected from subtidal region.

Appendix Table IID.--(continued).

	Stuart Island	Wood Point	Malikfik Bay	Cape Denbigh	Golovnin Lagoon	Rocky Point	Square Rock	Bluff Point	Safety Sound	Cape Nome	Sledge Island	Cape Woolley	King Island
PHYLUM ECHINODERMATA													
Class Asteroidea			x										
Crossaster papposus													x
Leptasterias cf. *L. hylodes*				x									
Leptasterias sp. §													x
Asterias amurensis										x			
Class Ophiuroidea			x										
PHYLUM UROCHORDATA													
Class Ascidiacea													
Pelonaia corrugata ¶				x									
Synoicum sp.										x			
Boltenia sp. §													x

§. Collected from subtidal region.
¶. Probably washed ashore from the sublittoral region.

Appendix III

Some benthic marine algae from the Pribilof Islands, Bering Sea: A Preliminary Annotated List

by

Natasha I. Calvin

ABSTRACT

Several collections of seaweeds at the Pribilof Islands were made in the subtidal zone, and one collection was made in the intertidal zone. The collections yielded 5 species of green algae, 12 species of brown algae, and 38 species of red algae. Thirty species are new records for the Pribilof Islands, and of these 6 are new records for the Bering Sea.

New species from the Pribilof Islands are: *Cladophora hutchinsiae* (Dillw.) Kutzing, *Urospora wormskioldii* (Mertens) Rosenvinge, *Codium ritteri* Setchell and Gardner, *Myrionema balticum* (Reinke) Fosl., *Desmarestia viridis* (Mueller) Lamouroux, *Cymathere triplicata* (Postels and Ruprecht) J. Agardh, *Laminaria dentigera* Kjellman, *Laminaria yezoensis* Miyabe, *Alaria fistulosa* Postels and Ruprecht, *Rhodochorton penicilliforme* (Kjellman) Rosenvinge, *Neodilsea americana* Abbott, *Neodilsea integra* (Kjellman) Zinova, *Rhodophysema elegans* var. *polystromatica* (Batters) Dixon, *Cryptonemia borealis* Kylin, *Callophyllis japonica* Okamura, *Cirrulicarpus* gmelini (Grunow) Tokida and Masaki, *Kallymenia oblongifructa* Setchell, *Turnerella mertensiana* (Postels and Ruprecht) Schmitz, *Rhodymenia pertusa* (Postels and Ruprecht) J. Agardh, *Halosaccion ramentaceum* (L.) J. Ag. f. *subsimplex* Kjellman, *Pleonosporium kobayashii* Okamura, *Odonthalia ochotensis* (Ruprecht) J. Agardh, *Pterosiphonia bipinnata* (Postels and Ruprecht) Falkenberg f. *bipinnata*, *Laingia aleutica* Wynne, *Phycodrys riggii* Gardner, *Tokidadendron ambigua* (Gardner) Wynne, *Tokidadendron bullata* (Gardner) Wynne, and *Zinovaea acanthocarpa* Wynne.

New species from the Bering Sea are *Neodilsea americana*, *Myrionema balticum*, *Rhodochorton penicilliforme*, *Rhodophysema elegans*, *Cryptonemia borealis*, and *Kallymenia oblongifructa*.

The most recent reported collection of seaweeds in the Pribilofs prior to this study was made in 1897.

INTRODUCTION

The Pribilof Islands lie in the Bering Sea, roughly 57°N, 170°W, about 6°N and 9°E of Amchitka Island of the Aleutian Islands.

William Setchell (1899), under the auspices of the U.S. Commission on Fur Seals and the Fur Seal Islands, summarized all records of marine algae in the Pribilofs up to that date, including those of Ruprecht (1851), (which concerned primarily the algae of the Okhotsk Sea, but made some mention of specimens from the Pribilofs) and Kastalsky, which were summarized by Postels and Ruprecht (1840). In addition, Setchell's summary included three other small collections: one made at his request in 1895 at St. Paul by Charles N. Townsend of the U.S. Fish Commission steamer ALBATROSS, another consisting of three jars of seaweeds collected at St. Paul Island and preserved in formalin by Messrs. Greeley and Snodgrass in the years 1896-1897 under direction of the Commissioner on Fur Seals and the Fur Seal Islands, David Starr Jordan, and a third containing a few species obtained by the Alaska Commercial Company about 1877. Apparently Setchell never went to the Pribilofs himself. Setchell's summary included 4 Chlorophyta, 10 Phaeophyta, and 21 Rhodophyta. Some of the algae were subtidal, having been dredged or found in the drift.

Collection of algae in Alaska has continued sporadically, but no collections have been reported from the Pribilof Islands since Setchell made his summary in 1899. The Harriman Alaska Expedition of 1899 and a series of algae-collecting expeditions sponsored by the University of British Columbia under the direction of Dr. Robert F. Scagel between 1959 and 1969 reached the Aleutian Islands, but did not continue to the Pribilofs.

Collections made at Amchitka Island in the Aleutians prior to the atomic bomb tests resulted in some new records and some valuable taxonomic work, chiefly that of Wynne (1970) on the Delesseriaceae.

The most comprehensive work on taxonomy and distribution of the Bering Sea marine algae to date is that of F. R. Kjellman (1889), which, though it does not include any records from the Pribilofs, is of inestimable help in solving the riddles of the algae of that region. The Language Services Branch, Office of International Fisheries, NMFS, translated this work from Swedish for purposes of the present study.

Setchell and Gardner (1903) in their summary of the marine algae of northwestern America, suggested that the Boreal Region of algal distribution be divided into an Upper and a Lower Region, with the Aleutian Islands as boundary, pointing out that many algal species, such as Nereocystis luetkeana, which extend up the Pacific Coast, stop at the Aleutian Islands and do not extend north into the Bering Sea. It is also true that some algal species which occur in the Bering Sea do not extend south of the Aleutians. It does appear that the Bering Sea has a distinct flora.

In August 1975 and June 1976 we had the opportunity to dive at the Pribilof Islands in conjunction with an intertidal study which was being conducted by the Auke Bay Laboratory of the National Marine Fisheries Service, NOAA, and which was related to effects of oil development in the area.

In this report I present a preliminary summary of seaweeds collected at those times. This report does not include information from intertidal transects.

METHODS

In August of 1975 divers from the Auke Bay Laboratory and the NOAA ship SURVEYOR made exploratory dives and species collections at several sites on St. Paul Island, St. George Island, and Otter Island, which, with the exception of some rock outcrops and reefs, constitute the Pribilof Islands. An intertidal collection was made at Zapadni Bay on St. George Island in August of 1975. In 1976 we collected quadrats subtidally at various depths on the three islands. The quadrat method assured collection of many smaller, inconspicuous species that would otherwise have been missed, provided exact depth and substrate information for each plant, and provided a means whereby divers who were not phycologically oriented could assist substantially with collecting. The quadrats were used as a species collection method only, and were placed subjectively by divers on the basis of apparent algal diversity rather than at any particular depth or substrate. We used 1/16 m^2 quadrats in areas of dense algal cover and 1/4 m^2 quadrats in areas of sparse cover. The samples were preserved in formalin and returned to the Auke Bay Laboratory for identification. Several expert taxonomists provided help with identifications (see Acknowledgements Section). Specimens are in the herbarium of the Auke Bay Laboratory, and numbers that follow the mention of some species refer to the Bering Sea section of that collection (BER).

SPECIES LIST

CHLOROPHYTA (green algae)

Order ULVALES

Family Monostromataceae

Monostroma fuscum var. *splendens* (Ruprecht) Rosenvinge, 1893

In the upper subtidal on the north side of St. George Island, 13 June 1976. Setchell (1899) reported a large number of specimens of this alga, as *Monostroma splendens*.

Order CLADOPHORALES

Family Cladophoraceae

Cladophora hutchinsiae (Dillw.) Kutzing, 1845

This green filamentous alga was reported from Bering Island (as *C. diffusa*) by Kjellman (1889), but has been reported from Alaska from only one collection, at Cook Inlet. We found it at Sea Lion Point on St. George Island at a depth of 10 ft on top of ice-scoured rocks on 9 June 1976.

Spongomorpha duriuscula var. *cartilaginea* (Ruprecht) Yamada

On rock in the upper subtidal, north side of St. George Island, 13 June 1976. Sterile. Width of filaments near upper part of plant about 450 μ; near base, 210 μ. Branching tends to be not alternate, but rather repeatedly on the same side. Setchell and Gardner (1903) report this plant from "St. Paul Island, Alaska", presumably in the Pribilofs, as *Cladophora Alaskana*.

Urospora wormskioldii (Mertens) Rosenvinge, 1893

Fairly common on upper surfaces of ice-scoured rocks. At Sea Lion Point (St. George Island) on 9 June 1976, cover of this alga on boulder tops at about 10 ft depth was about 30%. Many were fertile. Specimens from Otter Island collected 12 June 1976 had zoospores characteristically shaped for this species. Previously reported range California to the Aleutian Islands. Not previously reported from the Pribilofs.

Order COLDIALES

Family Codiaceae

Codium ritteri Setchell and Gardner, 1903

Reported on the basis of field notes only, not collected. Found at Rush Point on St. George Island, West of Zapadni Bay, and at Zapadni Bay near the seal observation site. Previous reported range of this plant is northern Southeast Alaska to the Aleutians. This is the first record from the Pribilof Islands.

PHAEOPHYTA (brown algae)

Order CHORDARIALES

Family Myrionemataceae

Myrionema balticum (Reinke) Fosl., 1894

On a Laminaria stipe in the upper subtidal zone at St. Paul Island on the east side of the southwest point reef. Fertile. Not previously reported north of California. Determined by Dr. Susan Loiseaux.

Order DESMARESTIALES

Family Desmarestiaceae

Desmarestia aculeata (?) (Linnaeus) Lamour

We saw occasional heavy patches of this alga on sand at 25 ft depth off the north side of St. George Island. Not collected. This species was reported from the Pribilofs by Setchell (1899).

Desmarestia viridis (Mueller) Lamouroux, 1813

We collected a small specimen at 15 ft on the southwest reef at Otter Island, 12 June 1976, and noted scattered clumps at Rush Point on St. George Island on 15 August 1976. This is the first record of this species from the Pribilofs. Kjellman does not mention it.

Order LAMINARIALES

Family Laminariaceae

Agarum cribrosum Bory, 1826

We noted this plant at several sites at about 20 to 30 ft depth, sometimes forming fairly extensive cover. It was reported in the Pribilofs by Setchell (1899) as *A. turneri*. His collection included only a fragment of a frond.

Cymathere triplicata (Postels & Ruprecht) J. Agardh, 1867

Both subtidal and lower intertidal: intertidal specimens were still juvenile in August 1975, and subtidal specimens were not full grown in June. We noted many juveniles in the shallow subtidal at Sea Lion Point, St. George. This species was previously reported in America only as far north as the Aleutians, though Kjellman (1889) reported sterile specimens washed ashore at Bering Island. This is the first record from the Pribilof Islands.

Laminaria dentigera Kjellman, 1889

Ubiquitous in the shallow subtidal area. Not reported by Setchell (1899). However, the type collection of this species is from Bering Island (Kjellman 1889).

Laminaria longipes Bory

Found often in the intertidal and upper subtidal areas. A 1/16 m^2 quadrat sample taken at Zapadni Point, St. Paul Island at about the 0 tide level yielded 89 blades of *L. longipes*, totalling 0.68 kg (=10.8/m^2) plus two *Alaria* sp. and two *L. dentigera*. Reported by Setchell from the collections of Messrs. Greeley and Snodgrass.

Laminaria yezoensis Miyabe, 1902

Found in the lower intertidal at Zapadni Bay, St. George Island, August 15, 1975. Sterile. We also observed a large subtidal patch of this plant at Sea Lion Point, St. George. Previously reported in Alaska as far north as the Aleutians. This is the first record from the Pribilof Islands.

Laminaria sp.

At Sea Lion Point, St. George, subtidal. August 14, 1975. Possibly L. groenlandica.

Thallasiophyllum clathrus (Gmelin) Postels and Ruprecht, 1840

Of major importance in the Pribilofs, this plant was found at almost every site sampled, occurring subtidally from about 10 to 25 ft, and reaching almost 50% cover of the bottom in some areas. Reported from the Pribilofs by Setchell.

Family Alariaceae

Alaria fistulosa Postels and Ruprecht, 1840

Forming floating beds offshore at most sites, this plant shows an affinity for the tops of boulders. A specimen collected at Otter Island on 12 June 1976 measured as follows: stipe, 29 cm; total length, 6.5 meters; width of blade 40 cm, weight, 1.35 kg. Its fertile sporophylls measured 30 cm long by 5 cm wide. Oddly, Setchell had no record of this conspicuous plant.

Alaria sp.

Probably more than one species of Alaria other than A. fistulosa occur both intertidally and subtidally on the islands. We have specimens from Zapadni Bay whose narrow sporophylls had only begun to appear on 15 August 1975. Kjellman reported a profusion of Alaria species from the Bering Sea. Setchell (1899) reported Alaria praelonga from the Pribilofs with the following comment: "A few specimens of an Alaria were collected by Mr. Townsend in

1895, which seem to belong to this species, although they have also the characters of A. angustata, A. crispa, and even of A. lanceolata; in fact, it is very difficult for the writer to determine how these four species differ essentially from one another." The present writer has similar problems.

Family Lessonicaceae

Nereocystis luetkeana (Mertens) Postels & Ruprecht, 1840

Setchell suggested that this species may occur in the Pribilofs, so it may be worth mentioning that we did not see it. I do not believe it occurs north of Unimak Pass in the Aleutian Islands.

RHODOPHYTA (red algae)

Order BANGIALES

Family Bangiaceae

Porphyra pseudolanceolata? Krishnamurthy, 1972

Intertidal at Zapadni Bay, St. George. One specimen is possibly male (BER101). These specimens have been sent to Dr. Thomas Mumford for determination.

Order NEMALIALES

Family Acrochaetiaceae

Acrochaetium cf. pectinatum

Epiphytic on an unidentified red alga (possibly Rhodymenia adnata) at 20 ft depth at St. Paul, 11 June 1976. Tetrasporic. (BER116).

Rhodochorton penicilliforme (Kjellman) Rosenvinge, 1893

At Otter Island, southwest reef, 12 June 1976 at 10 ft depth, on worm tubes, cells are approximately 13.2 μ wide, 33.4 μ long. Two tetraspores measured 28.8 μ long by 19.2 μ wide, and 24 μ long by 24 μ wide. Reported in Alaska from northern Southeast Alaska to northern Washington. Reported by Kjellman (1883) from the Arctic Sea (as R. mesocarpum f. penicilliformis), but not from the Bering Sea. This minute species would be easy to miss.

Order CRYPTONEMIALES

Family Dumontiaceae

Neodilsea americana Abbott, 1968

Three plants were collected from the north side of St. George Island, all subtidal. Blade thickness ranged from 136 μ to 210 μ. The plants are dark reddish brown and adhere firmly to the paper on drying. None are fertile. Not previously reported from the Bering Sea. Determined by I. Abbott. (BER 61, 77, 78)

Neodilsea integra (Kjellman) Zinova, 1961

Three plants were collected on the north side of St. George Island on 13 June 1976. One was epiphytic on the stipe of Thallasiophyllum. All were carposporic. All the plants are divided several times almost to the base. Thalli 136, 153, and 225 μ thick (BER 62, 79, 80). Reported from the Bering Sea by Kjellman (1889) as Sarcophyllis arctica. Not previously reported from the Pribilof Islands. Confirmed by I. Abbott.

Order CRYPTONEMIALES

Family Weeksiaceae

Constantinea rosa-marina (Gmelin) Postels & Ruprecht, 1840

Reported by Setchell from St. Paul in the Pribilofs, this red alga occurs as far south as southeast Alaska. It was one of the most ubiquitous algae in the upper subtidal zone at most sites visited, serving as substrate and cover for many invertebrates (most notably the terebellid worm, Nicolea zostericola and a snail, *Margarites* sp.). Sometimes almost completely covering boulder tops. Some small specimens were also found intertidally.

Family Peysonelliaceae

Rhodophysema elegans var. *polystromatica* (Batters) Dixon, 1964

Upper subtidal at St. Paul Island, epiphytic on a *Laminaria* stipe. The previously reported north Pacific limit of this plant is the San Juan Islands, Washington. Not reported from the Bering Sea. Identified through the courtesy of Darleen Masiak and Dr. John West.

Family Corallinaceae

Amphiroa setacea?

Subtidal on the southwest reef of Otter Island, 12 June 1976. Identification uncertain. Branching irregularly pectinate; intergenicula not winged; conceptacles on both axes and branches, one to several on a segment.

Crustose corallinaceae

The crustose corallines form a heavy cover in many areas in the upper subtidal zone. Some is extremely heavy and thick, and can be broken off in chunks. This type harbors animals underneath, such as soft bodied crabs, worms, clams, and black echiuroid worms.

Family Cryptonemiaceae

Cryptonemia borealis Kylin, 1925

This delicate red blade was found at several sites on St. George. All were sterile. Previously reported from Puget Sound south to California. (BER 55, 41, 44, 84).

Family Kallymeniaceae

Callophyllis japonica Okamura

Carposporic specimens were found on St. George Island at Rush Point and Sea Lion Point on 14 August 1975. This Japanese species has not been previously reported from Alaska or the eastern Pacific.

Callophyllis cristata (C. Agardh) Kuetz, 1849

This northern species has been reported from the Bering Sea to northern Washington. Reported from St. Lawrence Island, Bering Sea, by Kjellman (1889) as Euthora cristata. We found it ubiquitous in the subtidal zone. Most of our specimens come from around 20 ft depth. We found one carosporic specimen at Otter Island on 15 August 1975. This is the first record of this species from the Pribilof Islands.

Callophyllis adnata Okamura 1932

Found subtidally at Rush Point, St. George Island on 15 August 1975. This Japanese species has not been previously reported from Alaska or the eastern Pacific. Confirmed by I. Abbott. (BER 94)

Cirrulicarpus gmelini (Grunow) Tokida & Masaki, 1954

This red alga has been reported in Alaska only from the Aleutian Islands. It was a common species in the Pribilofs, occurring both in the low intertidal zone at Zapadni Bay and subtidally at several sites. We found it on rock, epiphytic on Thallasiophyllum stipes, and epizoic on tunicates. Plants found intertidally in August 1975 were sterile, but many subtidal plants were carposporic in June.

Kallymenia oblongifructa Setchell, 1912

Found at about 20 ft depth at all three islands. Often gregarious; more than twelve of these plants were found in one 1/4 m^2 sample quadrat on rock at 21 ft depth at St. Paul Island, 10 June 1976. None were fertile. All the plants were uniformly covered with fine papillate growths on both sides of the thallus, giving them the texture of fine sandpaper when dried. Previously reported from Cook Inlet, Alaska to northern California. Determined by I. Abbott.

Kallymenia sp.

One specimen of a *Kallymenia* bearing some resemblance to *K. reniformis* was collected 13 June 1976 on the north side of St. George Island. The blade is dark red and smooth, deeply cleft several times almost to the stipe, and about 225 to 262 μ thick.

Order GIGARTINALES

Family Nemastomataceae

Schizymenia borealis Abbott 1967

Several small specimens of a plant with gland cells, and which sticks to the paper on drying, are tentatively placed in this species. Found at about 20 ft depth.

Family Solieriaceae

Turnerella mertensiana (Postels & Ruprecht) Schmitz, 1889

This species was fairly common subtidally in both June and August. Two carposporic specimens were found at St. George Island in June 1976. Not previously reported from the Pribilofs. Determined by I. Abbott.

Family Gigartinaceae

Iridaea cordata (Turner) Bory var. *cordata*, 1826

One small specimen bleached and eroding was collected from the intertidal zone at Zapadni Bay, 15 August 1975. This species was reported from St. Paul Island by Setchell as *Iridaea laminarioides* f. *parvula*.

Order RHODYMENIALES

Family Rhodymeniaceae

Rhodymenia pertusa (Postels and Ruprecht) J. Agardh, 1851

Found on the northwest side of Otter Island, and the north side of St. George Island. Subtidal. Not previously reported from the Pribilof Islands.

Family Palmariaceae

Palmaria palmata f. typica (Kjellman) Guiry 1974

Found intertidally at Zapadni Bay, St. George Island on 15 August 1975, (BER 71) and subtidally at Sea Lion Point on the same date (BER 104, 96). Identification tentative.

Halosaccion ramentaceum (Linnaeus) J. Ag. f. *subsimplex* Kjellman 1883

We found several forms which seem referrable to *Halosaccion*. A narrow, flattened ribbon-like form, without proliferations, was found in the intertidal zone 15 August 1975 at Zapadni Bay. These plants had stalked tetrasporangia (BER 24, 91). A shallow subtidal form, also without proliferations, somewhat resembles a young *Palmaria palmata*, particularly in texture while fresh. Some of these are widened at the proximal end to 6 cm, while others are more linear with rounded tips. These plants were found at 10 to 12 ft, gregarious, and most have a tendency for the lower half of the plant to not adhere to the paper. This second group is only tentatively placed in *Halosaccion*. Setchell did not report this genus from the Pribilofs.

Family Ceramiaceae

Neoptilota asplenioides (Esper) Kylin, 1956

Fairly common subtidally. One specimen (BER 127) bears abundant carpogonial branches, often on opposite branches (both determinate and indeterminate) and on the thallus. Reported from St. Paul by Setchell as *Ptilota asplenioides*.

Pleonosporium kobayashii Okamura 1933

Tetrasporic specimens found at Otter Island southwest reef on 12 June 1976. Previously known only from Atka Island in the Aleutians. Confirmed by M.J. Wynne.

Ptilota serrata Kuetz

Common subtidally on rock or epiphytic. We found a tetrasporic specimen at St. George Island near the seal rookery at the west side of Zapadni Bay at 25 ft depth. Reported from St. Paul by Setchell as *Ptilota pectinata*.

An undescribed species in Ceramiaceae:

An alga which has not been described was found in the Pribilofs, scarce. This species was also found at Amchitka and is presently being considered by Dr. Michael J. Wynne. It is filamentous and heavily corticated, and clothed with fine, uniseriate, oppositely branched branchlets.

Family Rhodomelaceae

Odonthalia kamtchatica (Ruprecht) J. Ag., 1863

Found at several sites on rock at about 25 ft depth. Reported at Bering Island by Kjellman and at St. Paul Island by Ruprecht (and reported by Setchell).

Odonthalia ochotensis (Ruprecht) J. Ag.

Two carposporic specimens were found on 15 August 1975, one at Zapadni Bay at St. George, and one at Otter Island (BER 2, 16). Both specimens were at about 25 ft depth. This species is known from Japan; not previously reported from North America. Determined by M.J. Wynne.

Pterosiphonia bipinnata (Postels & Ruprecht) Falkenburg f. bipinnata, 1801

Found in the lower intertidal at Zapadni Bay, St. George, and upper subtidal (12 and 16 ft) at Sea Lion Point and Otter Island. Not previously reported from the Pribilof Islands, though it has been reported from the Arctic coast to southern California.

Family Delesseriaceae

Laingia aleutica Wynne 1970

Known only from the Aleutian Islands. We found several sterile specimens, some much eroded, at Sea Lion Point on St. George Island, and at the northwest side of Otter Island. This is the first report of this species from the Pribilofs. Determined by M. J. Wynne.

Membranoptera spinulosa (Ruprecht) Zionva, 1965

Subtidal, often epiphytic. First described from St. Paul Island in the Pribilofs, it is also found on the Bering Sea side of the Aleutians, in the Kuril Islands and on the Kamchatka Peninsula (Wynne 1970). We found tetrasporic specimens at the northwest side of Otter Island on 16 August 1975, and the north side of St. George on 13 June 1976, epiphytic on a Laminaria stipe.

Mikamiella ruprechtianum (Zinova) Wynne, 1977

We found several specimens of this plant at about 20 to 30 ft depth, on rock, at northwest Otter Island, on 16 August 1975. The type locality of this species is St. Paul Island. Determined by M.J. Wynne.

Myrogramme sp.

A number of small specimens (6 mm to 35 mm length) were collected at 21 ft on St. Paul Island, 10 June 1976, densely aggregated on worm tubes. Sterile. (BER 65, 90)

Phycodrys riggii Gardner, 1927

Many carposporic and tetrasporic specimens were found subtidally on the north side of St. George on 13 June 1976, and at Otter Island on 12 June. Sterile specimens were also found, two of them intertidal at Zapadni Bay, St. George. Known from the Aleutian Islands to northern southeast Alaska as well as Japan and the Kamchatka Peninsula; not previously reported from the Pribilof Islands.

Tokidadendron bullata (Gardner) Wynne, 1970

Sterile and tetrasporic specimens were found on the north side of St. George on 13 June 1976. Previously known from Sitka to the Aleutian Island; also found in Asia (Sakhalin and the Kuril Island). Not reported from the Pribilofs.

Yendonia crassifolia (Ruprecht) Kylin, 1935

A common subtidal alga in the Pribilofs, this species was reported by Setchell (as Delesseria crassifolia) in all collections available to him. The type locality of this species is St. Paul Island. We have two specimens from St. George with new vegetative blades perennating from old, carposporic blades. They were collected on 13 June 1976. (BER 133, 135).

Zinovaea acanthocarpa Wynne, 1970

We found one small specimen of this unique species at St. George on 13 June 1976 (BER 139). It has been found elsewhere only at Amchitka Island and Adak Island in the Aleutians (Wynne 1970). Not previously found in the Pribilofs. Collected by R.J. Ellis.

ACKNOWLEDGEMENTS FOR APPENDIX

Many divers assisted me with collections: from the Auke Bay Laboratory, Louis Barr and Robert Ellis dived in both 1975 and 1976. John MacKinnon dived in 1975 and made the collection of intertidal algae at Zapadni Bay, St. George. Divers from the NOAA ship *SURVEYOR* were, in 1975, Mark Howe and Hans Ramm, and in 1976, Steve Thorne, Pat McKeown, Gary Lagerlof, and William Harrigan.

The following people helped with identifications: Dr. Isabella Abbott, Dr. Susan Louiseaux, Darleen Masiak, Dr. John West, Dr. Michael J. Wynne, and Dr. Hiroshi Yabu, of Hokkaido University, Japan. Dr. Rita O'Clair identified the worm *Nicolea zostericola*.

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260	33	_delete_ † _F. berkeleyi_ = _Fabricia pacifica_, auctt. (Hobson and Banse, in press).

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260	19	"*Fabriciola berkeleyi* †" should be *Fabricia pacifica*
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260	33	delete "† *F*. *berkeleyi* = *Fabricia pacifica*, auctt. (Hobson and Banse, *in press*)."

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FINAL REPORT

Task Numbers A-27; B-9
Contract # 03-5-022-68
Research Unit #190

Study of Microbial Activity and Crude Oil-Microbial Interactions in the Waters and Sediments of Cook Inlet and the Beaufort Sea

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TABLE OF CONTENTS

SUMMARY STATEMENT

Page Number

A. Overview 424

B. General objectives of our studies 425

C. Relevance of this study to management decisions 425

D. Summary of major findings 425

E. Recommendations for future study 428

F. Acknowledgements 430

SECTION I. - COOK INLET

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development 432

II. Study areas 435

III. Methods 435

- A. Sampling procedures 435
- B. Relative microbial activity and percent respiration (mineralization) determinations 444
- C. Heterotrophic potential studies 446
- D. Direct cell counts 447
- E. Nitrogen fixation in sediments 448
- F. Nutrient analysis 449

IV. Results 451

- A. Gulf of Alaska (NEGOA) 451
- B. Cook Inlet 453

1. Relative microbial activity and respiration percentages in water. 453

2. Relative microbial activity and respiration percentages in sediments 458

3. Nitrogen fixation rates in sediments 458

C. Data storage 463

V. Discussion 467

A. Relative microbial activity and percent respiration 467

1. Comparison between activities in sediments and the overlying water columns 467

2. Seasonal differences 469

3. Geographical differences 470

B. Nitrogen fixation 476

1. Seasonal differences 476

2. Geographical differences 477

3. Significance of nitrogen fixation in overall productivity 478

VI. Conclusions 480

VII. Needs for further study 482

SECTION II. - BEAUFORT SEA

I. Summary of objectives, conclusions, and implications with respect of OCS oil and gas development 484

II. Study areas 487

III. Methods 492

A. Analysis of adenylate concentrations 492

IV. Results 494

A. Relative microbial activity and respiration percentages 494

1. Local variations 494

2. Geographical differences 495

3. Comparison of relative microbial activity in Beaufort Sea with other regions 501

4. The influence of freshwater input on cell concentrations and microbial activity 506

5. Seasonal variations 508

6. Comparison of microbial activity in sediments and in the water column 513

7. Adjustment of microbial populations to hydrocarbons in the environment 517

B. Rates of nitrogen fixation in Beaufort Sea sediments 517

C. Total adenylates in Elson Lagoon 523

D. Crude oil toxicity studies 523

E. Data storage 523

V. Discussion 525

A. Geographical differences 525

B. Seasonal differences 528

C. Role of microorganisms in ice 533

D. Effects of crude oil and the dispersant Corexit 9527 on microbial function 534

VI. Needs for further study 535

SECTION III. - NORTON SOUND

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development 537

II. Study area 539

III. Methods 539

IV. Results 539

A. Relative microbial activity 539

B. Nitrogen fixation 546

C. Crude oil effects studies 546

V. Discussion 551

VI. Needs for future research 553

SECTION IV. - EFFECTS STUDIES

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development 555

II. Study areas 559

III. Methods 559

- A. Sample collection and manipulation 559
- B. Procedures for assaying the acute effects of crude oil and/or Corexit 563
- C. Enzyme assays 565
- D. Redox potential and pH measurements 566
- E. Primary productivity rates 567
- F. Denitrification 567

IV. Results 569

- A. Short-term effects of crude oil and/or Corexit 9527 569
- B. Long-term effects of crude oil and/or Corexit 9527 599
  - 1. Effects on relative microbial activity in marine sediments 599
  - 2. Effects on respiration percentages 616
  - 3. Elson Lagoon study 617
  - 4. Glacier Bay study 621
  - 5. Effects on nitrogen fixation 621
  - 6. Effects on denitrification 627
  - 7. Effects on CO_2 and methane evolution rates 630
  - 8. Effects on total adenylates and energy charge 633
  - 9. Effects on bacterial concentration 638

10. Effects on sediment surface pH and Eh 638

11. Effects on sediments augmented with organic carbon 640

C. Seasonal observations of microbial function in the sediments and waters near Kasitsna Bay 651

V. Discussion 673

A. Short-term effects of crude oil and/or Corexit 673

1. Crude oil effects 673

2. Corexit effects 678

3. Effects of exposing natural microbial populations to both crude oil and Corexit 681

4. Short-term effects and management decisions 682

B. Long-term effects of crude oil on microbial function 682

1. Role of bacteria in marine systems - an overview 682

2. Role and relative importance of bacteria in the detrital food chain 684

3. Role and importance of bacteria in the mineralization process 687

4. Role and importance of microbial nitrogen fixation 688

5. Long-term crude oil effects on microbial function 691

a. Overview 691

b. Crude oil effects on the detrital food chain 692

c. Crude oil effects on mineralization and nitrogen fixation 695

d. Crude oil effects on sediment chemistry 698

e. Effects of "weathered" crude oil 699

f. Effects in sediments augmented with organic carbon 700

g. Effects on sediments collected from various locations 705

6. Long-term effects of the dispersant Corexit 9527 on microbial activities 706

C. Seasonal observations of microbial function near Kasitsna Bay 706

1. Carbon cycle 707

2. Nitrogen cycle 708

D. The importance of long-term crude oil effects relative to overall productivity 709

E. The relevance of our observations to actual spill conditions 711

F. Recommendations regarding the types of measurements that should be conducted during a major spill 713

SECTION V. CRUDE OIL WEATHERING

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development. 719

A. Objectives and background 719

B. Implications for off-shore oil and gas development 719

II. Methods 721

A. Extraction 722

B. Liquid column chromatography 723

C. Gas chromatographic analysis 723

D. Gas Chromatogram data reduction 724

E. Capillary gas chromatography mass spectrometry 726

III. Results 727

A. Time zero samples 727

B. Time one year samples 739

C. Sadie Cove oil/nutrient amended sediment experiments 757

IV. Discussion 760

A. Implication of hydrocarbon analyses 760

B. Interpretation of hydrocarbon data relative to microbial processes 763

V. Acknowledgements 766

Literature Cited 767

Glossary of Terms 780

SUMMARY STATEMENT

A. Overview

This report includes five major sections under the headings of "Cook Inlet", "Beaufort Sea", "Norton Sound", "Effects" and "Weathering". These sections are tied together by the appropriate cross-references but they were each designed to be complete statements of our work in each area. We have organized this report in this way so that those interested in just one area will be able to find all of the relevant information for that area within the appropriate section.

We understand that people with greatly varying scientific backgrounds may have use for the information contained in this report. With this in mind, we have attempted to explain our results and conclusions using nontechnical terms whenever possible. In order to aid in the understanding of the text, we have included a glossary of terms at the end of this report which we hope will be of help to those unfamiliar with some of the terms used.

We strongly urge all of those using this report to read parts B.1. and B.2. of the "Discussion" in Section IV. In this section, we have summarized the role of bacteria in the marine environment and the importance of microbial function in the overall productivity of marine ecosystems. Later in the same section, we describe the effects of crude oil on microbial function as it effects productivity in the marine environment.

B. General objectives of our studies

In each geographic area that we have studied, we have measured various microbial functions related to the overall productivity of the respective ecosystems. The types of variables that we have studied are outlined in detail in the main body of this report and will not be discussed in this section. In addition to collecting these baseline data, we have studied both long and short-term effects of crude oil and the dispersant Corexit 9527 on these and other functions. The results of these studies are presented in Section IV.

C. Relevance of this study to management decisions

Whenever possible, we have tried to relate our findings to management decisions concerning the production and transport of crude oil. Most of the statements are presented in part I of each major section. We have also outlined the potential adverse effects of crude oil on the overall productivity of Alaskan marine ecosystems in part D of the "Discussion" in Section IV.

D. Summary of major findings

1. We have found that crude oil alters microbial function in marine sediments. This altered function will have three major impacts on normal biological activity. (1) It will reduce overall productivity by interfering with the normal flow of food through the detrital food chain. Recent estimates show that 50-80% of food available to all animals present is ultimately derived from this source. (2) Crude oil will interfere with the processes that convert the nitrogen and phosphorous in organic material into inorganic forms which are required for plant growth. Without these inorganic nutrients, plants can not produce the new organic material required to feed the animals present. (3) Crude

oil changes microbial activity in the sediments so that the chemical environment of the sediment surface is changed. It seems quite likely that these changes will remain long after the initial crude oil toxicity has abated. These changes could greatly alter the normal recruitment of animals back into the impacted area.

2. The specific changes that we have observed in marine sediments which are exposed to crude oil are the following:

a. Reduction in nitrogen fixation and deitrification rates
b. Reduction in microbial biomass production
c. Reduced phosphatase activity
d. Reduced enzymatic activity in those enzymes that hydrolyze structural polysaccharides.
e. Increased enzymatic activity in those enzymes that hydrolyze storage polysaccharides.
f. Decreased total biomass including bacterial biomass
g. Increased methane CO_2 concentrations
h. Decreased redox potentials and increased surface hydrogen ion concentrations
i. Decreased infaunal borrowing
j. Increased accumulation of detrital material on the sediment surface

3. When we studied the effects of various concentrations of fresh crude oil on several of the above functions, it was found that a maximum effect could be observed at concentrations as low as 1 ppt v/v. These studies also showed that the most sensitive indicators of fresh crude oil perturbation were microbial biomass production, nitrogen fixation and redox potential changes.

4. When we studied the effects of "weathered" crude oil on the same processes, we found that nitrogen fixation rates were not affected but that denitrification rates and microbial biosyntheses were significantly affected by the presence of "weathered" crude oil. From this we have concluded that the types and intensity of the effect will depend, in part, on the composition of the hydrocarbon perturbing the system.

5. The season at which the perturbation occurs is probably much less important than the qualitative and quantitative characteristics of the crude oil introduced into the sediments.

6. We also conducted time course experiments which were designed to establish how long it takes for some of these changes to take place after fresh crude oil is introduced into the sediments. In many cases, altered function was observed within the first two days exposure. If one assumes that it takes 2-3 days for crude oil from a spill to be transported into the sediments in nearshore environments, it would take a total of 4-5 days from the start of a spill until changes in microbial function would be observed. This fact should be kept in mind by those concerned with planning environmental impact studies following an oil spill. If the initial measurements are not made within the above time frame, there would be little chance of obtaining information about "prespill" conditions prior to the onset of changes in response to crude oil perturbation.

7. In our estimation, the most vulnerable environment in Alaskan marine systems is the soft-fine grained sediments such as those found in the St. Georges Basin in the southern Bering Sea, Shelikof Strait, and the major bays of Cook Inlet. These are the regions for which we would predict the greatest long-term perturbation in the case of a large scale

oil spill. This would be particularly true in the soft sediments landward of the barrier islands near the major rivers in the Beaufort Sea.

8. We suspect from the length of exposure time it takes for microbial function in Beaufort marine sediments to change when perturbed with crude oil, that these marine sediments would be impacted for a much longer period of time than in more temperate marine sediments. There is also indirect evidence that suggests marine organisms depend heavily on detritus as their major food source during the winter months. If these assumptions are true, the Beaufort Sea would appear to be particularly susceptible to crude oil perturbation.

9. In summary, the results of our studies suggest that every precaution should be taken to keep crude oil out of marine sediments. In areas where it is suspected that the detrital food chain in marine sediments is particularly important to the overall productivity of a region, special care must be taken to prevent spills from occurring and in the case of an actual spill, cleanup procedures which would cause the crude oil to be driven into the sediments should be avoided. This would also hold for those areas in which susceptible microbial functions are known to be of particular significance to the overall productivity.

E. Recommendations for future study and planning

We have listed recommended directions for future research under part VII of each section. These ideas are summarized below.

1. A concerted effort should be made in the area of planning for a coordinated research effort in the event of a major spill in Alaskan marine waters. In the "Executive Summary" of The Tsesis Oil Spill final report, it was concluded that in future environmental impact studies, a contingency research plan be formulated before an actual spill occurs.

It was also recommended that funding be identified to support research in the event of an oil spill. We strongly recommend that OMPA/NOAA establish such a plan for Alaskan marine environments. This plan should address the problems of what types of studies should be conducted, sampling strategies and logistical requirements. We further recommend that the NOAA laboratory at Kasitsna Bay be seriously considered as the field analytical laboratory for the initial phases of this work. We would also like to stress that it would be very important to initiate these studies as soon as possible after a spill occurs. If changes in microbial function are to be studied, it would be advantageous if the initial observations could be made within 5 days of the initial impact. This would allow us to establish baseline measurements of relatively non-altered function prior to the onset of anticipated changes.

2. At the present time there is very little information about microbial processes in Prince William Sound, the Chukchi Sea and the Bering Sea. There is information which indicates that the detrital food chain in the St. Georges Basin (southern Bering Sea) could be the main route through which organic carbon is made available to all marine animals in this area (Iverson, et al., 1979). We strongly recommend that studies of key microbial functions be conducted in the St. Georges Basin so the potential impact of crude oil on the productivity of this extremely important fishery could be evaluated. In addition, we recommend that crude oil impact studies be conducted on sediments collected in this region.

3. Our preliminary work in the Beaufort Sea suggests that inshore regions may be very susceptible to crude oil perturbation for extended periods of time. We recommend that studies of crude oil effects be initiated in regions landward of the barrier islands which are near the major rivers of the North Slope.

4. In the last 1.5 years, we have learned a great deal about the impact of crude oil on microbial function in marine sediments; however, there are still a number of areas that should be explored. (a) Although we have made observations that strongly suggest that the detrital food chain is interrupted by the presence of crude oil, we have not documented this by direct observations. We suggest that a study be initiated which would address this problem.(b) At the end of this investigation, we have established that certain microbial functions are altered by the presence of crude oil in marine sediments; however, much more information must be collected before adequate predictive capabilities can be established. What is needed is a long-term study on the effects of various concentrations of different crude oil fractions on key microbial processes. This would enable us to predict the impact of a given oil on these processes under actual spill conditions. In order to make these predictions, we would require qualitative and quantitative information about the crude oil content of marine sediments in the impacted area.

F. Acknowledgements

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COOK INLET

Section I

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development.

A. Objectives

The first three cruises (October 1976, April 1977 and November 1977) were designed to produce baseline data on the rates at which microorganisms utilize and mineralize organic nutrients and the rates at which they fix atmospheric nitrogen. During the November 1977, April 1978 and April 1979 cruises, we were to make the above mentioned observations in addition to measuring the short-term (acute) effects of Cook Inlet crude oil on these processes. These observations were to be made in coordination with hydrocarbon chemists to determine if microbial populations adjusted to the presence of crude oil (April 1978 and 1979). These investigations were coordinated with the studies conducted by another group of microbiologists (Atlas RU #29).

In addition to these measurements, we were to collect data on the abiotic variables of salinity, temperature and the biotic variables of inorganic nutrient concentrations of NH_4^+, NO_3^-, and PO_4^{-3}. These latter variables are mainly microbially controlled.

Our studies were designed to determine when and where the impact of crude oil would be the greatest in Cook Inlet. Our conclusions based on the observations made during these five cruises and other information collected during the existence of RU #190 are listed below.

B. Conclusions and Implications

During the course of our studies, we have determined that crude oil has an adverse short and long-term effects on microbial processes in both Arctic and sub-Arctic waters and sediments of the Alaskan Continental Shelf. The dispersant Corexit 9527 was also observed to have an adverse short-term effect on heterotrophic activity. The net result of the impact of crude oil is to reduce overall productivity in the perturbed region. These results are presented and analyzed in Section IV of this report. What we will present in this section is a summary of our recommendations relative to gas and oil development in Cook Inlet using all of the information available to us from all of our OCSEAP studies to date. In this discussion, we have also included potential impacts to Shelikof Strait which is essentially an extension of Cook Inlet.

1. Oil spilled in the Upper Cook Inlet would become associated with the large concentration of suspended matter in these waters. The crude oil would then be transported into the sediments of Kamishak Bay and the Shelikof Strait. (This conclusion stems from the work of Feeley and Cline on suspended matter in Cook Inlet).
2. Oil spilled in the Lower Cook Inlet would most probably drift into Kamishak Bay where it could become incorporated with the sediments of that region (Miller and Allen, 1976). If crude oil became dispersed throughout the water column, it could migrate into the sediments of Shelikof Strait.
3. Our studies have shown that microorganisms within the water column are initially stressed by the presence of either crude oil

or the dispersant Corexit 9527. This perturbation would not have a significant impact on the overall productivity of the affected area except under certain conditions; i.e., where crude oil remains in contact with the water column for an extended period of time or where crude oil becomes associated with suspended matter in the water column.

4. The major impact would undoubtedly occur in cases where the oil became incorporated into the sediments. We strongly recommend any procedures which would prevent crude oil from becoming incorporated into the sediments. Conversely, any procedures used in crude oil production and transport or in crude oil spill control which increases the chances of crude oil becoming incorporated into the sediments should be avoided.

5. The presence of crude oil in Alaskan marine sediments will adversely affect the overall productivity of the impacted region in several ways. We have shown that crude oil reduces the rate at which bacteria mineralize organic nutrients by over 50% under certain conditions. This reduces the rate at which inorganic nutrients (nitrogen, phosphorous, and sulfur) are released from organic matter. This could, in turn, reduce primary productivity rates. It also greatly reduces the rate at which atmospheric nitrogen is fixed in a form that can be used by all organisms. In addition, crude oil appears to interfere with the transfer of organic nutrients from bacteria to the balance of the detrital food chain.

6. The long-term impact of crude oil perturbation will be much greater in soft sediments than in the water column or long high-

energy coastlines. The concentration of crude oil in marine sediments required to produce measurable alterations in microbial function will be 1 ppt or less.

7. In our judgement, the areas where crude oil would have the greatest impact would be in Kachemak Bay. Impacts on Kamishak Bay and Shelikof Strait would be of lesser but still significant importance.

8. Since the impact of crude oil in marine sediments probably remains for several years, the season during which a spill occurs is probably of little direct importance to the severity of the impact. There could be secondary effects such as the frequency of storms which would tend to drive crude oil into the sediments.

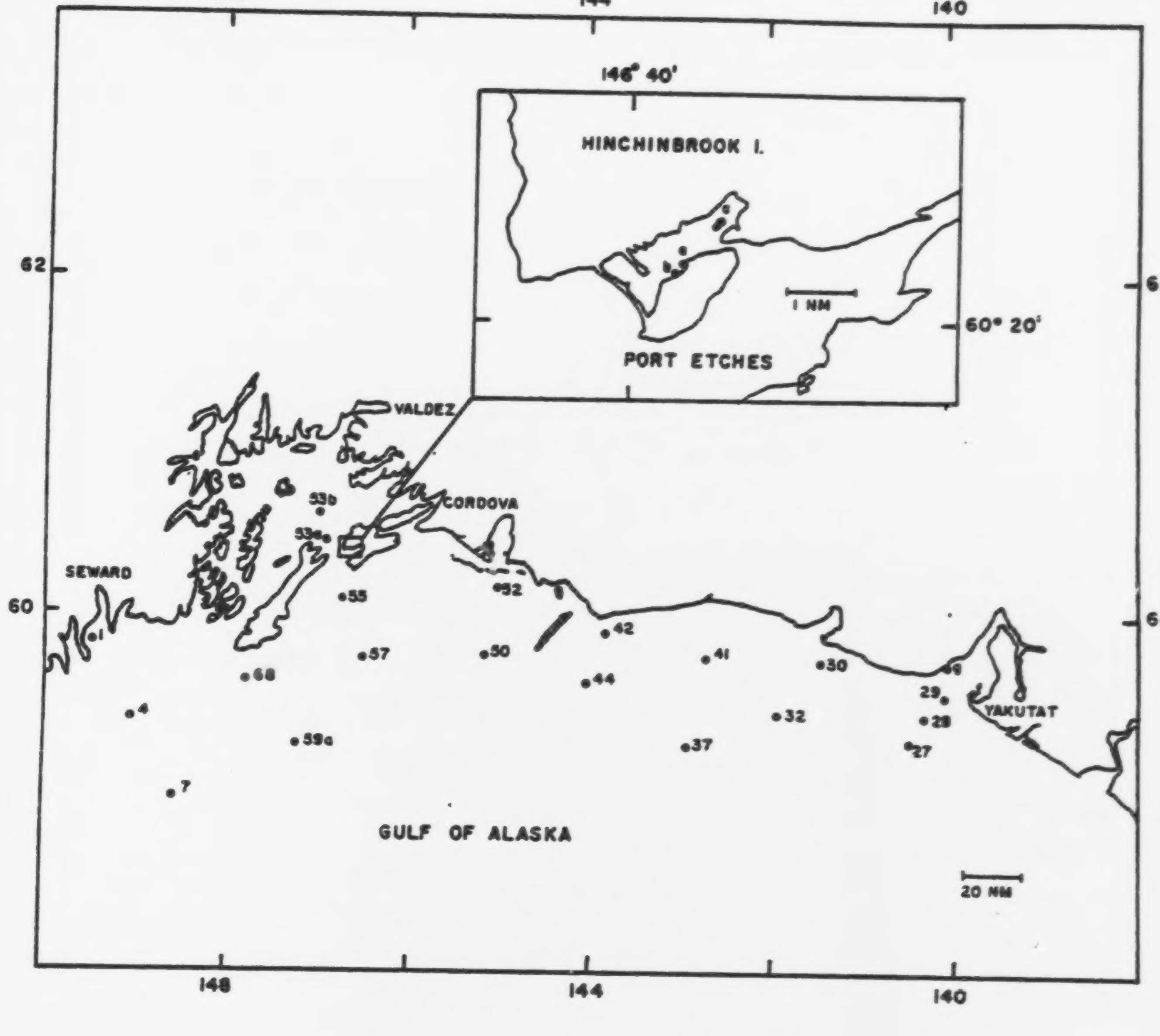
II. Study areas

We conducted one cruise in the North East Gulf of Alaska (NEGOA). The stations at which samples were collected are shown in Fig. 1. During the course of our Cook Inlet studies, we have participated in five major cruises: October 1976, April 1977, November 1977, April 1978 and April 1979 . The stations at which samples were collected are illustrated in Figures 2-8. During the first cruise, 37 water and 12 sediment samples were collected; during the second, 44 water and 12 sediment samples were collected; during the third, 60 water and 20 sediment samples were collected; during the fourth cruise, 83 water and 30 sediment samples were collected; and during the last cruise, 49 water and 14 sediments were collected.

III. Methods

A. Sampling procedures

The water samples were taken in sterile Niskin plastic water sample bags fitted on Niskin "butterfly" water samplers. In most



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HINCHINBROOK I.
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60° 20'
PORT ETCHES
62
60
VALDEZ
CORDOVA
SEWARD
53b
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41
30
57
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68
44
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YAKUTAT
GULF OF ALASKA
20 NM

Figure 1. Stations sampled in the Gulf of Alaska during the March 1976 cruise.

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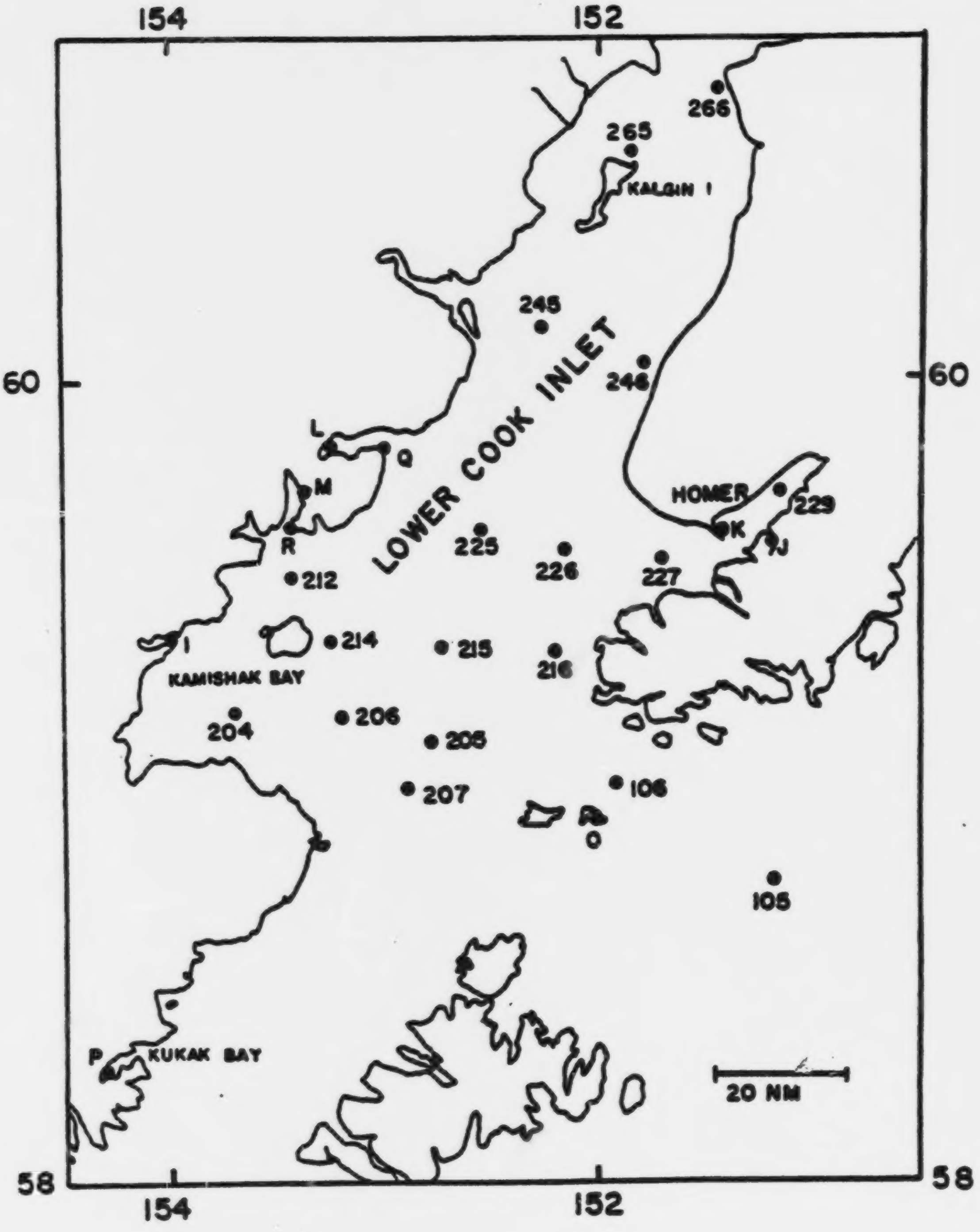
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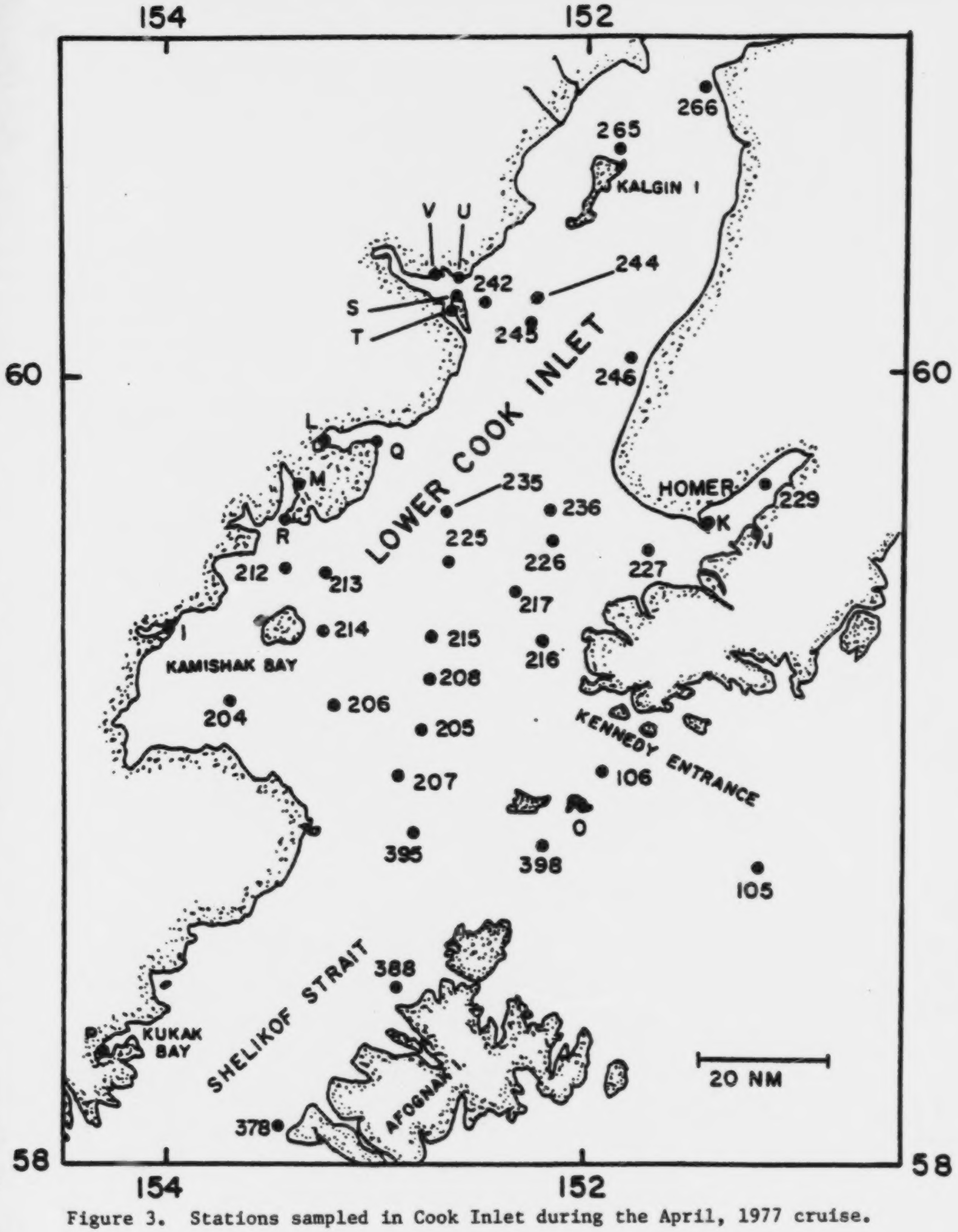
Figure 2. Stations sampled in Cook Inlet during the October 1976 cruise.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KAMISHAK BAY
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KENNEDY ENTRANCE
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SHELIKOF STRAIT
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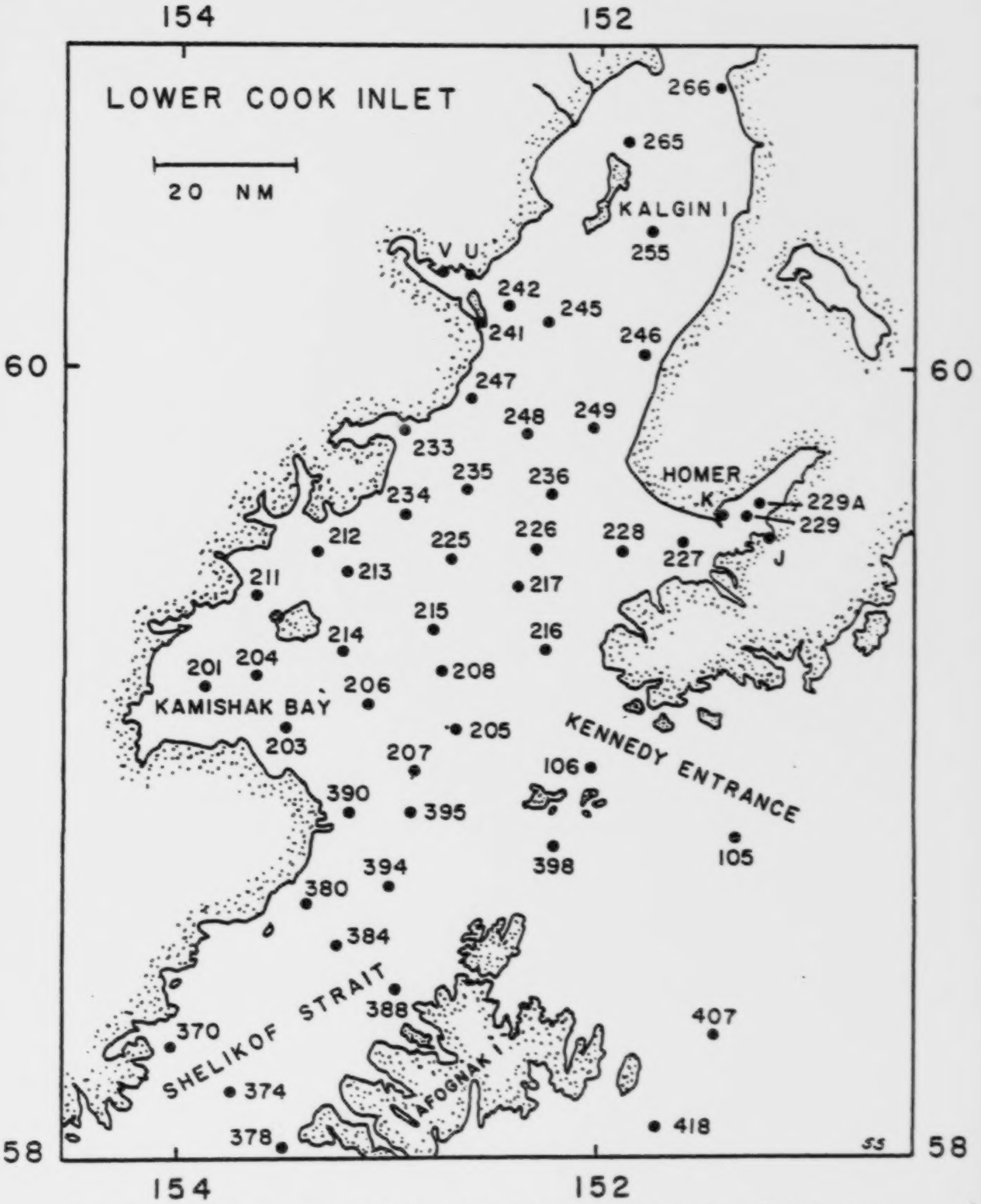
KUKAK BAY
20 NM
AFOGNAK I.
378

Figure 3. Stations sampled in Cook Inlet during the April, 1977 cruise.



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KENNEDY ENTRANCE
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AFOGNAK I
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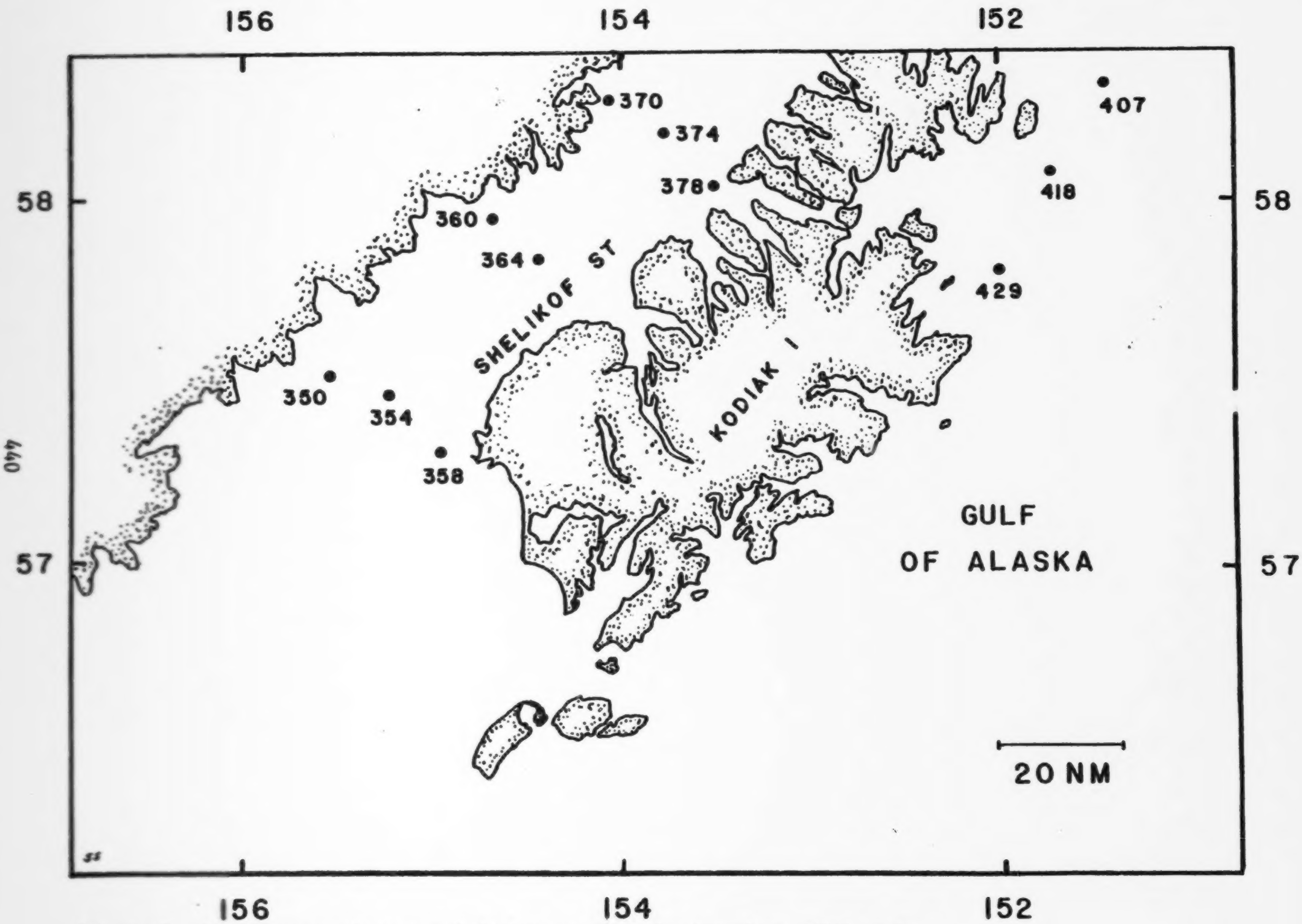
Figure 4. Stations sampled in Cook Inlet during the November 1977 cruise.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KODIAK I
GULF OF ALASKA
20 NM

Figure 5. Stations sampled near Kodiak Is. during the November 1977 cruise.

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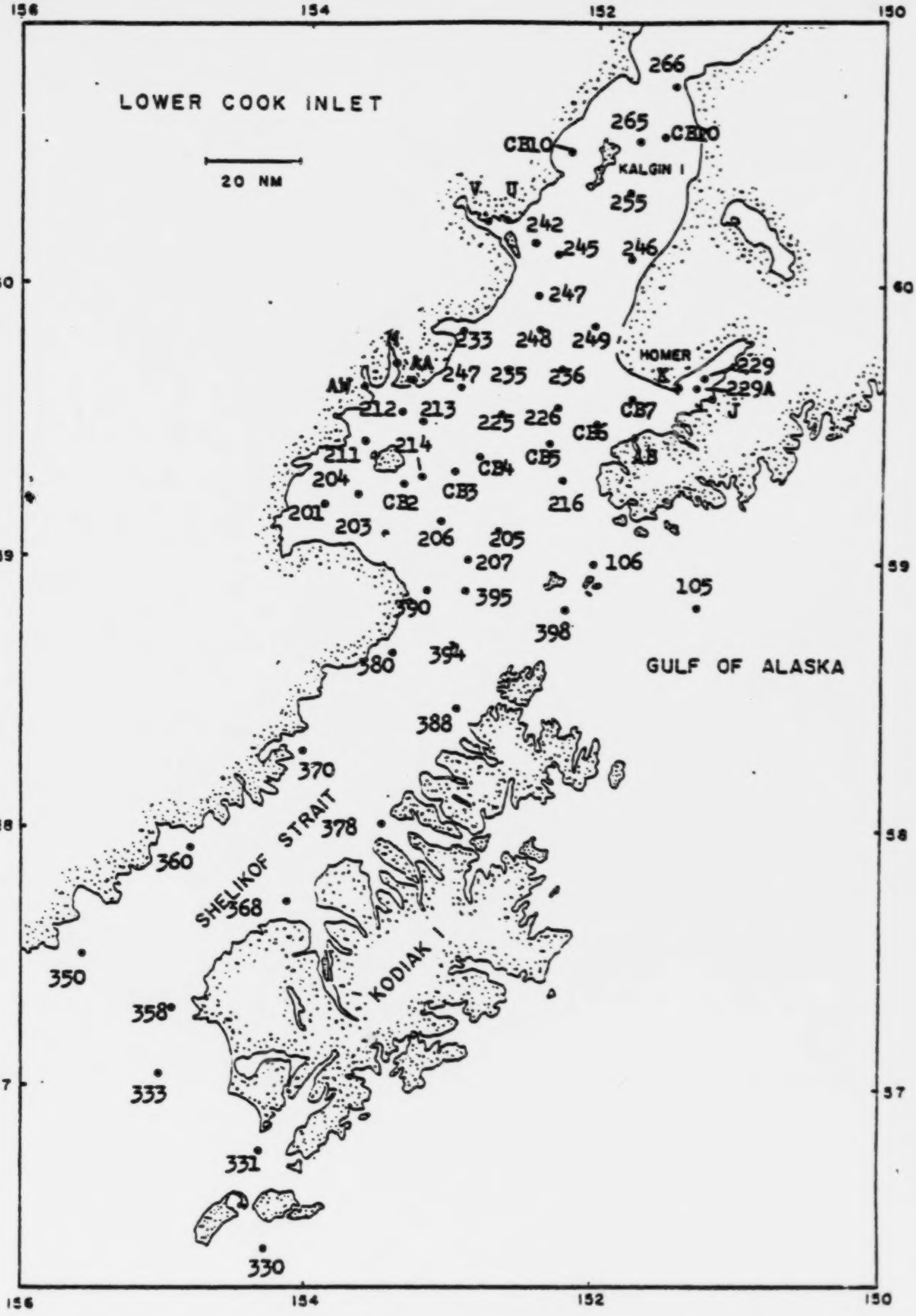
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AB
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CB4
CB2
CB3
216
201
203
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59
207
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105
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398
380
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GULF OF ALASKA
388
370
58
378
360
SHELIKOF STRAIT
368
KODIAK I
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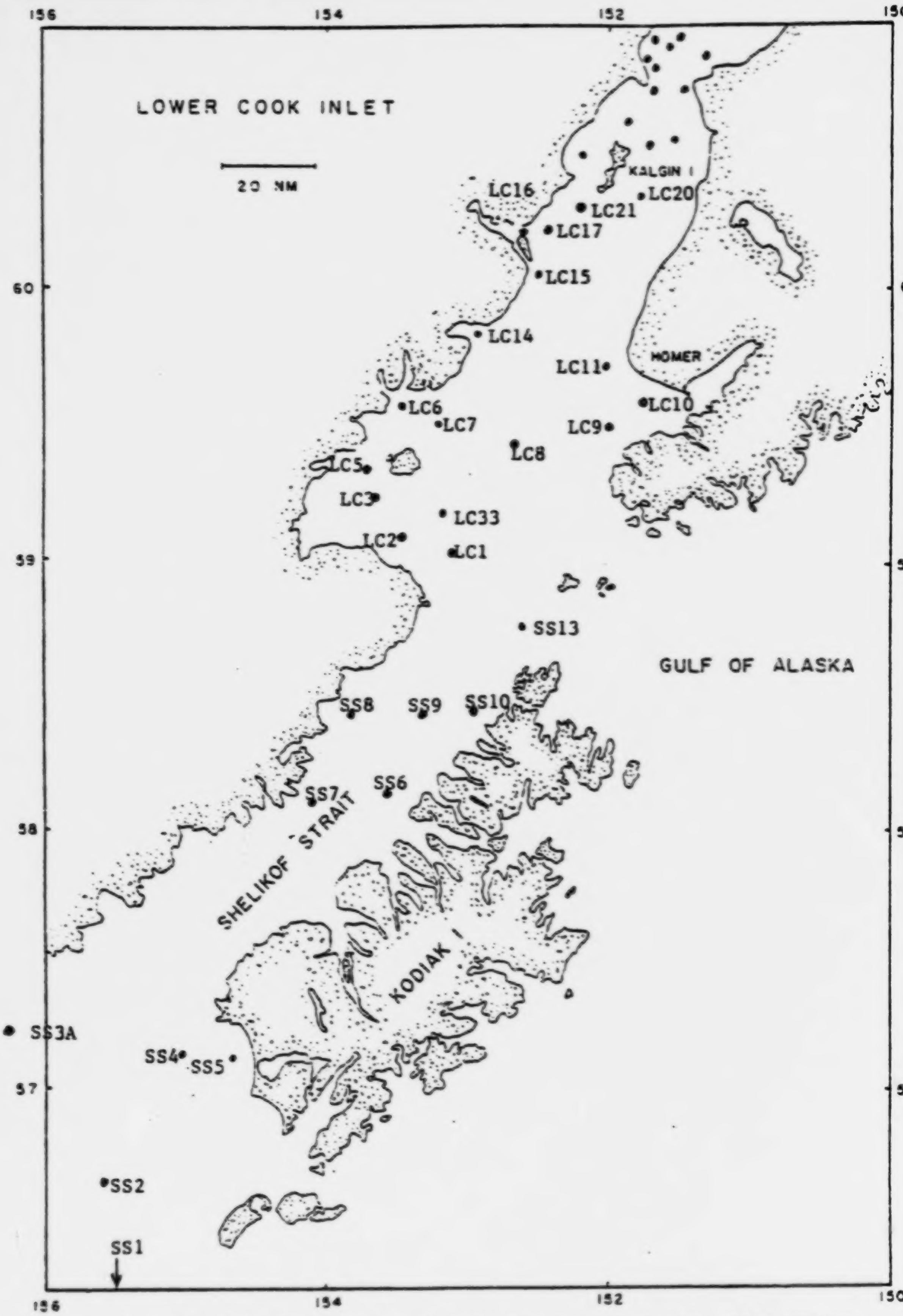
Figure 6. Location of stations sampled during the April 1978 Cook Inlet cruise.


156
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LOWER COOK INLET
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HOMER
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LC5
LC3
LC33
LC2
LC1
59
SS13
GULF OF ALASKA
SS8
SS9
SS10
SS7
SS6
58
SHELIKOF STRAIT
KODIAK I
SS3A
SS4
SS5
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SS2
SS1
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152
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Figure 7. Stations sampled in Cook Inlet during the April 1979 cruise.

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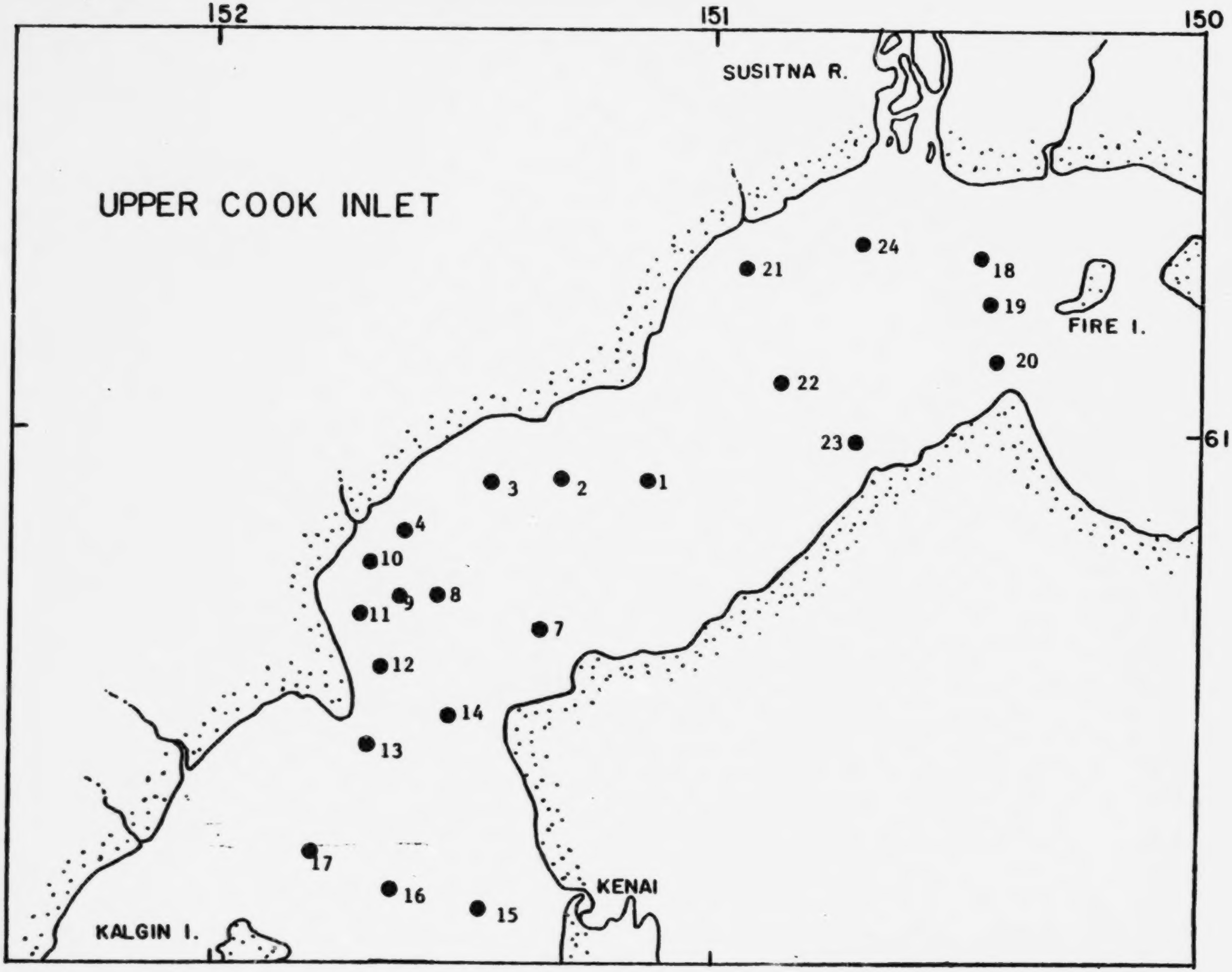
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SUSITNA R.
UPPER COOK INLET
FIRE I.
KENAI
KALGIN I.
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Figure 8. Stations sampled in the Upper Cook Inlet during the April 1979 cruise.

cases, water samples were taken within one meter of the surface. Once the water sample was taken, it was placed in an ice chest for storage and transported back to the laboratory for analysis. The analyses of microbial activity were initiated within two hours after sampling was terminated. During transport and storage, the samples were kept at or below the in situ temperature.

The majority of the sediment samples in the earlier cruises were taken with a Sutar-Van Veen bottom grab with some samples taken with a Shipek grab in coarse sediments. During the last two cruises, most of the sediments were taken from a one m^2 box corer. When practical, the top 2 cm were collected for use. The sediments that were collected from a small boat near shore, were sampled using a Kahl mud snapper. Water associated with the sample was used to make a sediment slurry for use in the subsequent analysis.

B. Relative microbial activity and percent respiration (mineralization) determinations.

The procedure used in these studies involved adding a U-^{14}C compound to replicate 10 ml subsamples which were contained in 50 ml serum bottles. After addition of subsamples, the serum bottles were sealed with rubber serum bottle caps fitted with plastic rod and cup assemblies (Kontes Glass Co., Vineland, M. J: K-882320) containing 25 x 50 mm strips of fluted Whatman #1 chromatography paper. In the sediment samples, a 10.0 ml subsample was diluted 1,000 times (v/v) with a 32 o/oo (w/v) solution of sterile artificial seawater. Ten ml subsamples of the sediment slurry were dried and weighed to determine the dry weights. These dry weights were used to calculate the observed uptake rates in terms of grams dry weight of sediment.

The samples were incubated in the dark within 0.5 C of the in situ temperature. After the incubation period, the bottles were injected through the septum with 0.2 ml of 5N H_2SO_4 in order to stop the reaction and release the $^{14}CO_2$. After the addition of the acid, 0.15 ml of the CO_2 adsorbent, β-phenethylamine, was injected onto the filter paper. The serum bottles were then shaken on a rotary shaker at 200 rpm for at least 45 minutes at room temperature to facilitate the adsorption of CO_2. The filter papers containing the $^{14}CO_2$ were removed from the cup assemblies and added to scintillation vials containing 10 ml of toluene based scintillation fluor (Omifluor, New England Nuclear).

The subsamples were filtered through a 0.45 μm membrane filter (Millipore). The trapped cells on the filter were washed with three 10 ml portions of seawater at 0-3 C. The filters were dried and then added to scintillation vials containing 10 ml of the above mentioned fluor. The vials were counted in a Beckman model LS-100 liquid scintillation counter located in our laboratory at Oregon State University.

During these studies, C^{14} labeled glucose with specific activity of approximately 300 mCi/mM and a final concentration of about 4 μg/liter was used in sediment samples and glutamic acid with a specific activity of about 230 mCi/mM and a final concentration of about 5.5 μg/liter was used in water samples.

Triplicate subsamples were analyzed for each sample. The channels ratio method for determining counting efficiencies was used. The observed CPM was converted to DPM before the mean value was calculated. The percent respiration (mineralization) was calculated by dividing the amount of labeled carbon taken up by the cells (both cell and CO_2 radioactivity) and multiplying this ratio by 100. All samples were incubated in the dark at a temperature within 0.5 C of the *in situ* temperature.

C. Heterotrophic potential studies

The technique used in these studies were basically those of Hobbie and Crawford (1969) as further modified by Harrison, Wright, and Morita (1971). This procedure involves the addition of different concentrations of U-^{14}C labeled substrate to identical subsamples. The procedures used to process the samples was identical to that described above. The only difference in these two methods is in the number of substrate concentrations used. In the first method, only one substrate concentration is used, in this method, four concentrations were used for each sample. Comparison of these two techniques was made by us (Griffiths et al., 1977) and it was concluded that both could be used to measure relative microbial activity.

Kinetic parameters were calculated using the following modification Lineweaver-Burk equation:

$$\frac{Cut}{c} = \frac{K_t + S_n}{V_{max}} + \frac{A}{V_{max}}$$

where c = radioactivity assimulated plus that respired as $^{14}CO_2$ by the heterotrophic population in disintegrations/min; S_n = the natural substrate concentration in μg/liter; A = the added substrate in μg/liter; C = 2.2 x 10^6 dpm/μCi of ^{14}C; μ = amount of ^{14}C labeled substrate added/sample bottle; t = incubation time in hours; V_{max} = the maximum velocity of uptake in μg x liter $^{-1}$ x h^{-1}; and K_t = the transport constant in μg/liter. From this equation can also be calculated the time

(T_t) in hours required by the natural microbial population to utilize the natural substrate in seawater sample. For the derivation of this equation and the assumptions on which it is based, see Wright and Hobbie (1966).

D. Direct Cell Counts

Ten ml of seawater was fixed in the field laboratory by adding it to 1.0 ml of membrane (0.45 μm) filtered formaldehyde (37%). The vials containing the fixed water samples were sealed and stored until they could be counted in our laboratory at Oregon State University. In the sediment studies, the final dilution of the sediments in the heterotrophic potential studies was used and treated the same as the seawater samples.

From 5 to 17 ml of sample were filtered through a 0.2 μm Nuclepore filter. When a relatively high number of organisms was present, the samples were diluted with membrane filtered artificial seawater. The number of organisms per field was kept within countable limits and the volume filtered was kept above 5 ml. Controls were run using filtered artificial seawater with all of the reagents used in the staining and mounting procedure. These counts were not more than 5% of the those found in the samples and were considered insignificant.

The staining procedure used was that of Zimmermann and Meyer-Reil (1974). This procedure involves staining the cells trapped on the membrane filter with acridine orange and then destaining with isopropyl alcohol. The membranes were dried and mounts on microscopic slides with a mounting medium of cinnamaldehyde and eugenol (2:1).

The bacterial cells were counted using a Zeiss IV Fl epifluorescence condenser microscope with filters KP 500, KP 490, FT 150, and LB 520. The eyepiece was used KpT W 12.5 x and the objective was plan 100 x. Approximately 50 restriction fields were counted per sample. Representative fields were counted from the center of the membrane filter to the outside edge of the filtration circle.

Only bodies with distinct fluorescence (either orange or green), clear outline and recognizable bacterial shape were counted as being bacterial cells.

E. Nitrogen fixation in sediments

Nitrogen fixation in the sediments was determined in the field by using the acetylene reduction method (Stewart et al, 1967). Ten ml subsamples of sediment were added to respective 50 ml serum bottles: one control and two duplicate samples were used for each analysis. After the bottles were sealed with a rubber stopper, the samples were gassed for one minute with helium at a flow rate of 10 cc/sec. Ten ml of acetylene was then added to each bottle and the bottles were allowed to incubate for 24 hr before being terminated with one ml of saturated $HgCl_2$ solution. The controls were treated in the same way before incubation and were used to determine the amount of ethylene that was released abiotically. After the incubation was terminated, the tops of the rubber stoppers were sealed with silicone cement. The bottles were kept at or below 4 C until they could be assayed for ethylene in our laboratory at Oregon State University. The analysis for ethylene was made on a Hewlett Packard model 5830A gas chromatograph. The column used was 1.9 meter of 1/8" stainless steel tubing packed with Porapak R (80-100 mesh) and the column temperature was 40 C. The carrier gas was nitrogen flowing at a rate of 29 cc/min. The resulting levels of ethylene were normalized using incubation times and gram dry weight conversions. All rates were calculated in terms of ng nitrogen fixed per gram dry weight of sediment per hour using a factor of 0.33 to convert the amount of ethylene measured to the theoretical amount of nitrogen fixed.

F. Nutrient analysis

1. Water sample nutrients

a. Frozen samples were thawed in a warm water bath and then aspirated into a four channel Technicon Autoanalyzer system. The samples were subdivided with a stream divider into four sample flows which were used to analyze ammonia, phosphate, nitrate and nitrite concentrations.

b. The total concentrations of nitrate and nitrite were determined following the procedures of Callaway et al. (1972). The following flow modifications were made to this procedure: sample, 0.8 cc/min; DDW diluture, 1.2 cc/min; ammonium chloride, 1.0 cc/min; sulfanilamide, 0.1 cc/min; N-1-naphylethylene 0.1 cc/min. The debubbler before the cadmium column is pumped out of the system (1.0 cc/min) with the remaining water forced through the cadmium column.

c. The nitrite concentration was determined using the same chemistry as the above analysis except there is no ammonium chloride, cadmium column, DDW diluter, or first air bubble. A 2.3 cm cc/min sample tube was used.

d. The phosphate concentration determinations were made using the method of Calloway et al. (1972) without modification.

e. The ammonium ion concentration was made using the technique of Head (1971).

2. Sediment samples

a. Sediment samples were thawed in a warm water bath, mixed and then centrifuged for 30 min at 0°C and 8000 x g.

b. Five to fifteen ml of the supernatant was removed and used in the nutrient analysis. Approximately 20 ml of the diluted sediment water was placed into quartz tubes with 0.3 ml of H_2O_2. These samples were then treated with UV light for 4 hours.

c. Soluble oxidizable nitrogen was determined as nitrate on the Auto-analyzer. The remainder of the diluted sediment water was diluted further for the ammonia determination.

d. When H_2S was present, approximately 0.15 ml of a 2% $CuSO_4$ solution was added to remove sulfide ions from solution which would interfere with the nutrient assays.

e. The total carbon content of the sediment was determined by the following procedure. A subsample of the sediment was treated with HCl to remove all traces of inorganic carbon. The sediment was centrifuged and the supernatant removed. The sediment was dried and combusted using the technique of Pella and Columbo (1973).

G. Statistical analysis

Differences in sampling and treatment mean values were analyzed using Student's "T" test. Unless otherwise stated, a critical value of $p < 0.05$ was used to define a "significant" difference.

Linear relationships were analyzed using the "least-squares" technique for regression and the "product-moment" technique for correlation coefficient.

IV. Results

A. Gulf of Alaska (NEGOA)

In March, 1976, we participated in an oceanographic cruise on board the NOAA ship Discoverer. Twenty-seven water and twenty sediment samples were taken at the stations shown in Figure 1. The kinetics of glutamic acid uptake were measured along with percent respiration and bacterial concentrations in both water and sediment samples (Table 1). From the kinetic data we were able to calculate the maximum potential rate of substrate uptake (V_{max}) which, in turn, can be used as an index of relative microbial activity. In the water samples, the mean V_{max} value was 1.4 ng x l^{-1} x h^{-1} which was approximately 1/2 of the mean value observed one month later in Beaufort Sea waters (Table 1) and much lower than that observed in Beaufort summer water samples. The mean V_{max} value observed in the sediments however was much higher than any V_{max} means that were observed in Beaufort Sea or in the Cook Inlet. This unusually high activity was not however reflected in an unusually high bacterial concentration. In fact, the mean bacterial concentration in NEGOA was only 50% higher than that observed in the winter Beaufort Sea sediments even though the relative microbial activity was almost 100 times as high.

There were no significant geographical patterns observed for relative microbial activity in the NEGOA water samples; however,

Table 1. Data summary of the average values measured during 1975-1976 field studies. (*) Average values calculated with one example excluded; a value which we consider more typical.

Factor	Units	Beaufort Sea Summer 1975		Beaufort Sea Winter 1976		Beaufort Sea Summer 1976		Lower Cook Inlet Fall 1976		Gulf of Alaska Winter 1976	
		Ave.	Range	Ave.	Range	Ave.	Range	Ave.	Range	Ave.	Range
V_{max} (Offshore water)	ng/liter/hr	40	4 to 118	3.1	0.2 to 14	21	0.4 to 85	28 (9.3)*	0.2 to 405	1.4	0.3 to 3.4
V_{max} (beach water)	ng/liter/hr	-	- -	-	- -	-	- -	84	0.7 to 404	63.0	9.7 to 113
$V_{max} \times 10^{-1}$ (sediments)	ng/g dry wt/h	5.2	0.2 to 17	0.5	0.04 to 1.8	6.9	0.2 to 64	7.1 (2.0)*	0.2 to 63	45	2 to 103
Percent Respiration (water)	%	59	44 to 76	85	52 to 100	46	20 to 59	58	40 to 78	72	53 to 93
Percent Respiration (sediments)	%	43	32 to 71	45	35 to 87	28	14 to 35	46	38 to 53	44	30 to 72
Number of bacteria $\times 10^5$ (seawater)	cells/ml	4.5	0.1 to 11.9	1.5	0.8 to 2.7	3.7	1.6 to 2.7	4.2	0.2 to 16.5	1.9	1.2 to 2.7
Number of bacteria $\times 10^8$ (sediments)	cells/g dry wt	6.3	0.1 to 41.1	10	0,5 to 19	106	24 to 267	41	4 to 130	15	0.1 to 31
Sample temperature (surface water)	°C	1.2	-0.8 to 3.2	-1.9	-2.0 to -1.5	0.3	-1.3 to 1.8	8.3	-1.5 to 12	3.8	2.0 to 5.0
Sample salinity (surface water)	o/oo	20.5	9.0 to 26.5	24	17 to 29	12.3	5.1 to 20.5	23.8	20.5 to 27.5	31.9	30.7 to 35.5

there was a pattern of relative microbial activity in the sediments that corresponded to areas of high concentrations of detritus feeding benthic organisms (H. Feder, personal communication).

B. Cook Inlet

Between October 1976 and April 1979, we participated in five major cruises in the Cook Inlet. The first three taken in October 1976, April 1977 and November 1977 were designed primarily as a means of collecting baseline information on microbial function in this area. The remaining two cruises in April 1978 and 1979 were designed as coordinated study to be conducted with chemical oceanographers.

1. Relative microbial activity and respiration percentages in water.

a. Relative microbial activity was, on the average, lower in the waters collected during the November 1977 cruise and higher in the April 1979 waters than samples collected during any of the other cruises (Table 2). Both glucose and glutamic acid uptake rates (relative microbial activity) were significantly higher ($p<0.05$) in water samples collected in April 1979 than at any other time. The differences between the mean relative microbial activity values observed during the November 1977 cruise and the April 1978 and 1979 cruises were statistically significant but the differences between the mean observed during the November 1977 cruise and the other two cruises were not. The ratio of glucose to glutamic acid uptake was highest in the April 1978 and 1979 water samples. The differences seen in the mean percent respiration ratios were not statistically significant.

Table 2. Relative microbial activity of Cook Inlet waters, excluding Homer Boat Basin and Shelikof Strait.

		Oct. 1976		Apr. 1977		Nov. 1977		Apr. 1978		Apr. 1979	
Activity	Units	$\bar{x}$	range	$\bar{x}$	range	$\bar{x}$	range	$\bar{x}$	range	$\bar{x}$	range
Glutamate uptake (T)	ng/l/hr	8.0	0.2-53	9.7	0.5-665	5.3	0.4-61	7.3	0.1-75	18.9	0.8-58
Glucose uptake (S)	ng/l/hr	3.0	0.2-33	1.6	0.2-12	0.8	0.2-6.6	4.5	0.1-20	11.9	0.7-72
Ratio S/T	-	0.38		0.16		0.15		0.62		0.63	
Glutamate Respiration	%	58	40-78	48	27-70	55	32-70	50	32-71	54	35-70
Glucose Respiration	%	28	10-84	41	5-45	31	14-89	29	14-53	35	13-48

b. A series of experiments were conducted which were designed to determine if the waters near the beach (in the surf zone) showed higher relative microbial activities than those observed further off-shore (10 meters off-shore from the surf zone). In every case where this was measured in Cook Inlet and in the Gulf of Alaska, the relative microbial activity was highest in the surf zone.

c. With the exception of the April 1979 cruise, patterns of relative microbial activity were similar in all Cook Inlet cruises. These patterns reflect differences in the microbial communities associated with the major water masses in this region. These water masses are roughly defined by surface water salinities. These patterns are best illustrated in the data collected during the April and November 1977 cruises (Fig. 9). There are essentially two major water masses in Cook Inlet. One is open ocean water which is characterized by the higher salinities found in the southeastern portion of the Inlet. The other water mass originating from the Upper Cook Inlet, is characterized by relatively low salinities.

As shown in Figure 10, the relative microbial activities in the northern waters are highest observed in this region, the lowest values were observed to the south and east of the inlet and in open ocean waters. Intermediate values were observed in samples collected along the western side of the inlet. These measurements and the respiration percentages listed below were determined using labeled glutamic acid.

Consistent respiration percentage patterns were also seen when the results of the data collected were compared from various

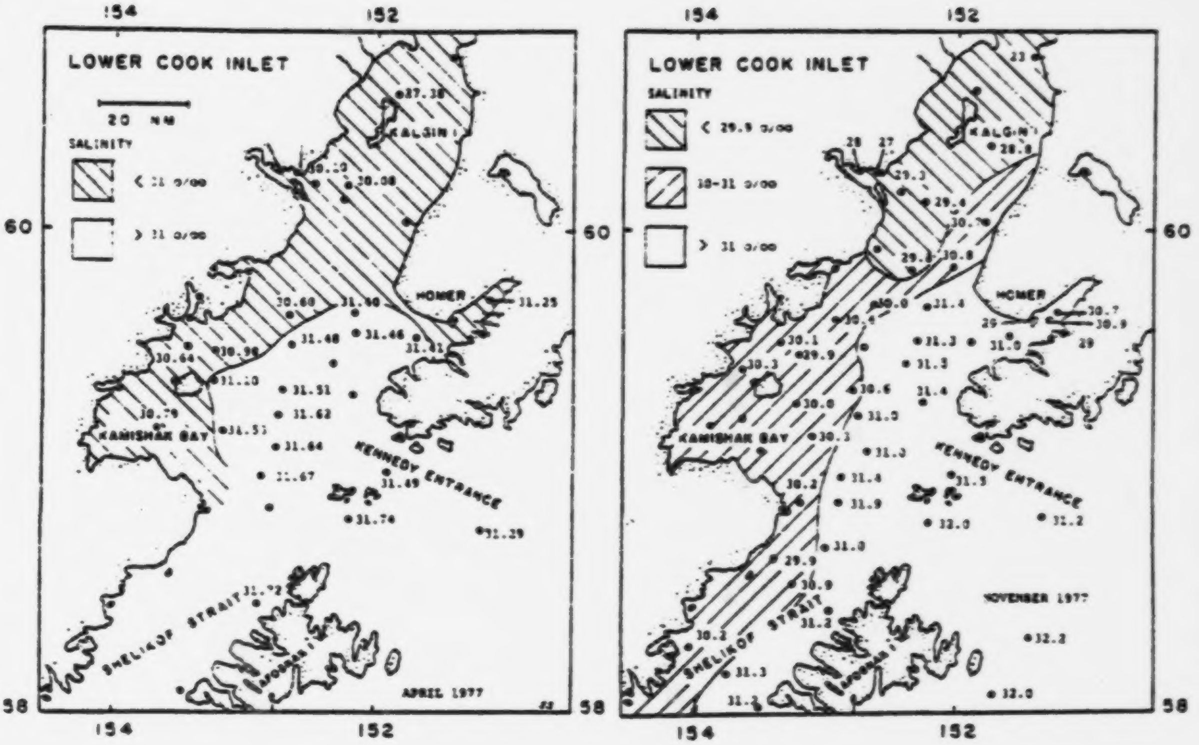
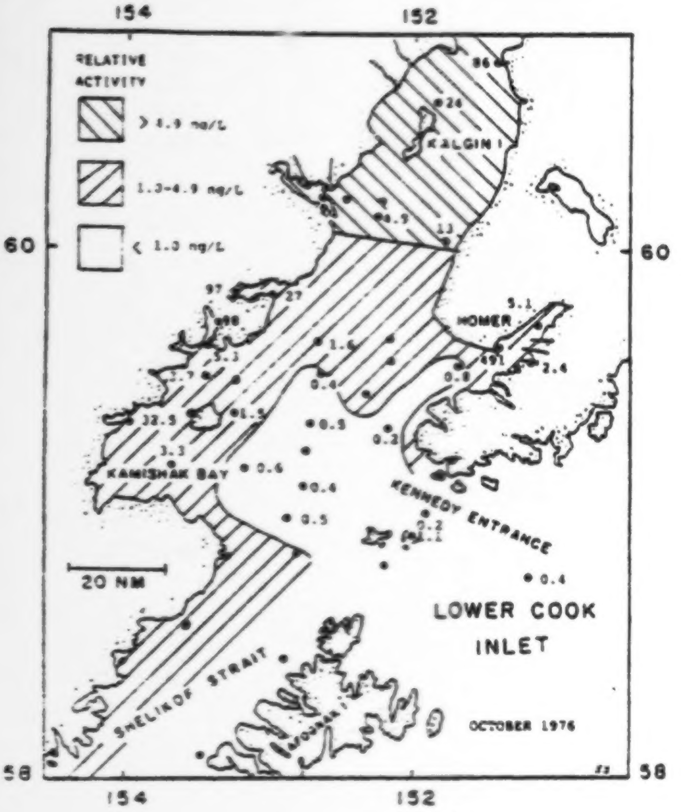
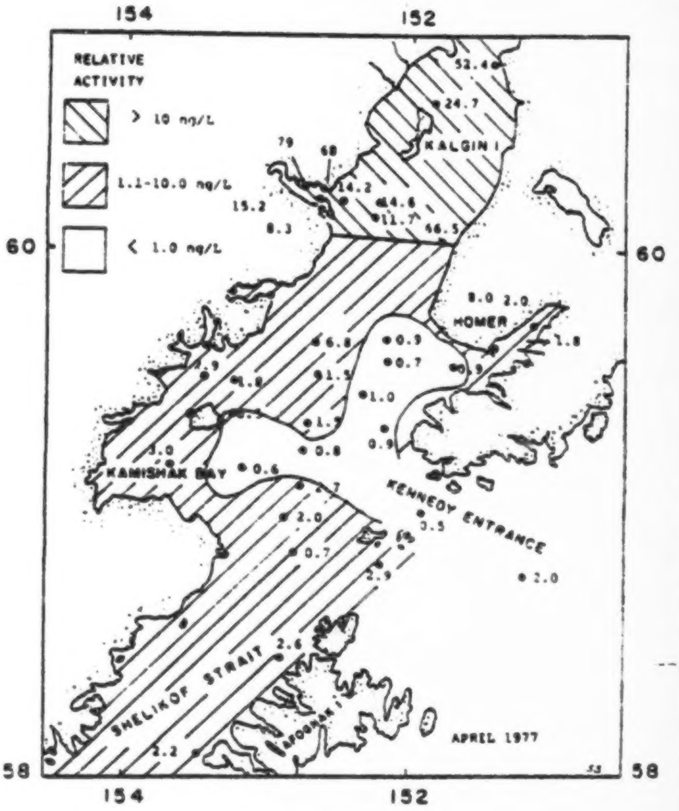

154
152
LOWER COOK INLET
20 NM
SALINITY
< 31 o/oo
> 31 o/oo
60
58
KALGIN I
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
27.38
30.10
30.08
30.60
31.40
31.25
31.48
31.46
31.41
30.64
30.98
31.10
31.51
31.62
30.79
31.53
31.64
31.67
31.49
31.74
31.29
31.72
APRIL 1977
154
152
LOWER COOK INLET
SALINITY
< 29.9 o/oo
30-31 o/oo
> 31 o/oo
60
58
KALGIN I
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
23
28.8
28
27
29.3
29.4
30.7
29.8
30.8
30.0
31.4
30.4
30.7
30.9
29
29
31.3
31.0
30.1
29.9
31.3
30.3
30.6
31.4
30.0
31.3
30.3
31.3
31.4
31.5
30.2
31.9
32.0
31.2
31.0
29.9
30.9
NOVEMBER 1977
31.2
30.2
32.2
31.3
32.0
31.2

Figure 9. Surface salinities in Cook Inlet during the April and November 1977 cruises.


154
152
RELATIVE ACTIVITY
> 4.9 ng/L
1.3-4.9 ng/L
< 1.0 ng/L
60
58
KALGIN I
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
20 NM
LOWER COOK INLET
SHELIKOF STRAIT
AFOGNAK I
OCTOBER 1976


154
152
RELATIVE ACTIVITY
> 10 ng/L
1.1-10.0 ng/L
< 1.0 ng/L
60
58
KALGIN I
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
APRIL 1977

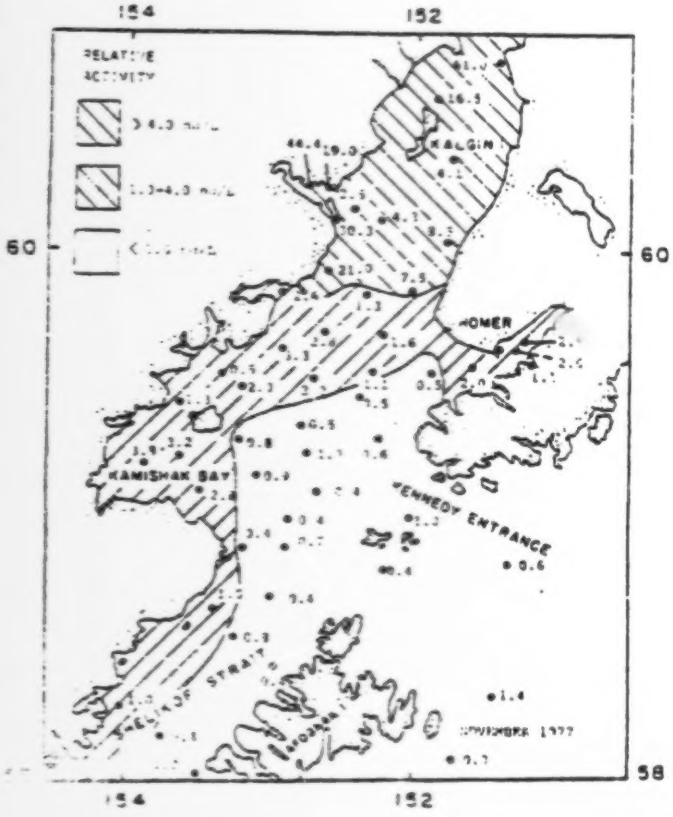

154
152
RELATIVE ACTIVITY
> 4.3 ng/L
1.3-4.3 ng/L
60
58
KALGIN
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
NOVEMBER 1977

Figure 10. Water masses in Cook Inlet characterized by relative activities during three sampling periods. These measurements were made using glutamic acid.

cruises (Fig. 11). The values in the area near Kalgin Island and Tuxedni Bay are very low ranging from 31 to 40%. Contours of increasing values run in lines which run diagonally from the northeast to the southwest. Intermediate values are found along these contours in the center of the inlet and the highest values are found in the southeastern portion of the inlet and in the open seawater.

During the April 1979 cruise, the levels of relative microbial activity were unusually high (Table 2). These high activity levels seemed to mask the patterns of microbial activity that we had seen in the past cruises. We interpret these high values as being a response by the microbial population to a spring phytoplankton bloom. This phenomenon altered patterns of relative microbial activity more than it did respiration percentages.

2. Relative microbial activity and respiration percentages in sediments.

Both the relative microbial activity and respiration percentages observed in the sediments collected during all five cruises showed little variation regardless of the season in which they were sampled (Table 3). In general, the highest rates of microbial activity were observed in the major bays within the Inlet. These values were consistently higher than those observed in the Shelikof Strait. These geographical patterns were best illustrated during the April and November 1977 cruises when the most comprehensive sediment sampling took place (Figs. 12 and 13).

3. Nitrogen fixation rates in sediments.

With the exception of the October, 1976 cruise, measurable nitrogen fixation rates were observed in sediments collected during

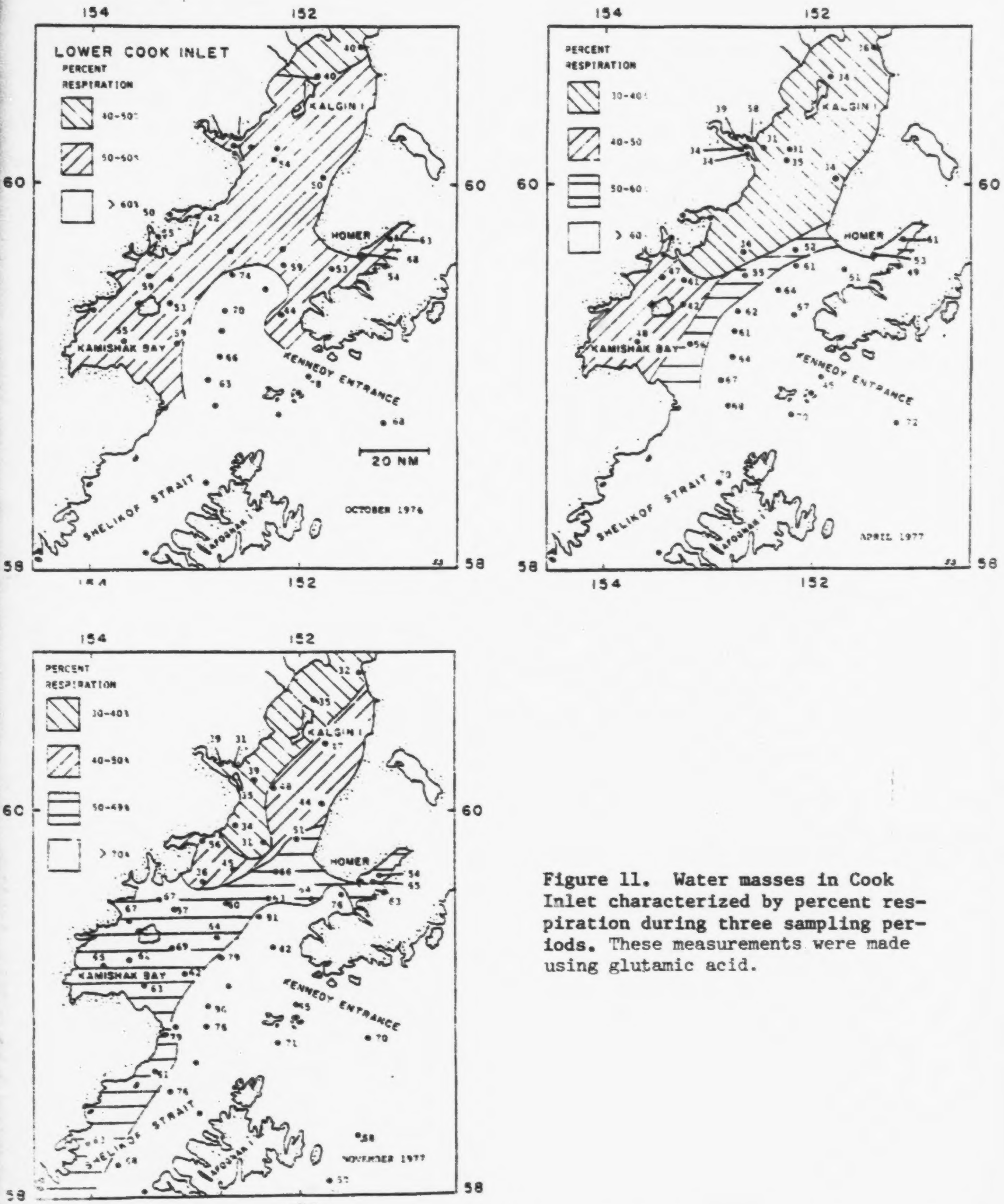

LOWER COOK INLET
PERCENT RESPIRATION
40-50%
50-60%
> 60%
KALGIN I
HOMER
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
20 NM
OCTOBER 1976
APRIL 1977
30-40%
NOVEMBER 1977
> 70%
154
152
60
58

Figure 11. Water masses in Cook Inlet characterized by percent respiration during three sampling periods. These measurements were made using glutamic acid.

Table 3. Relative microbial activity of Cook Inlet sediments, excluding Shelikof Strait.

Activity	Units	Oct. 1976		Apr. 1977		Nov. 1977		Apr. 1978		Apr. 1979	
		$\bar{Y}$	range	$\bar{Y}$	range	$\bar{Y}$	range	$\bar{Y}$	range	$\bar{Y}$	range
Glut. uptake	ng/g/hr	131	30-360	240	80-370	103	20-252	131	10-600	208	72-550
Glucose uptake	ng/g/hr	3.3	0.9-7.1	8.3	2-18.4	6.1	0.7-23.3	6.5	0.3-38.4	5.2	0.9-17.8
Glut. Resp.	%	46	38-53	39	40-49	49	43-57	46	31-61	46	39-51
Glucose Resp.	%	28	20-43	23	16-36	22	13-40	26	10-33	22	14-28

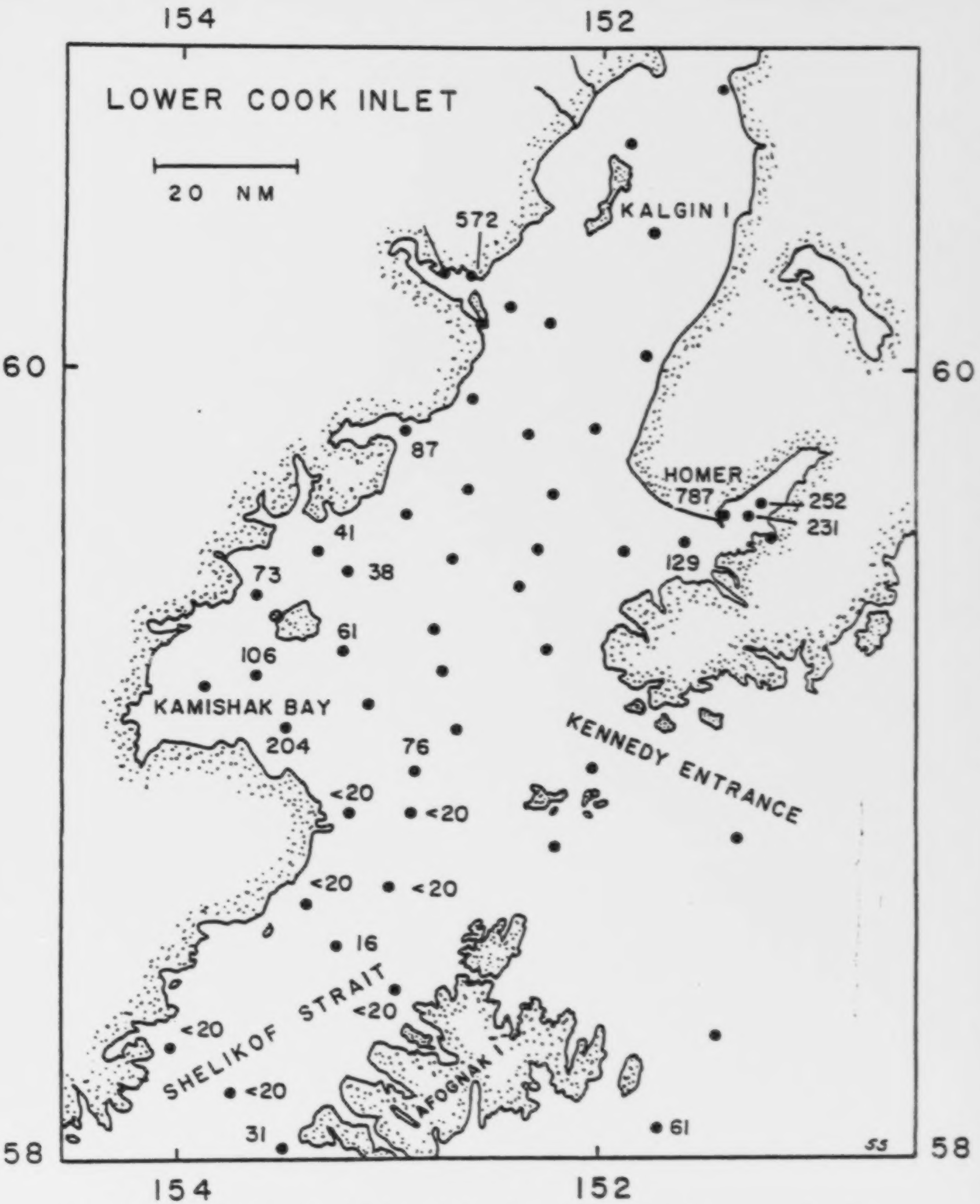

154
152
LOWER COOK INLET
20 NM
572
KALGIN I
60
60
87
HOMER
787
252
231
41
38
129
73
61
106
KAMISHAK BAY
204
76
KENNEDY ENTRANCE
<20
<20
<20
<20
16
SHELIKOF STRAIT
<20
<20
<20
AFOGNAK I
31
61
SS
58
58
154
152

Figure 12. Relative microbial activity in sediment samples measured using glutamic acid during the November 1977 cruise. Units are ng/ g dry wt./ h.

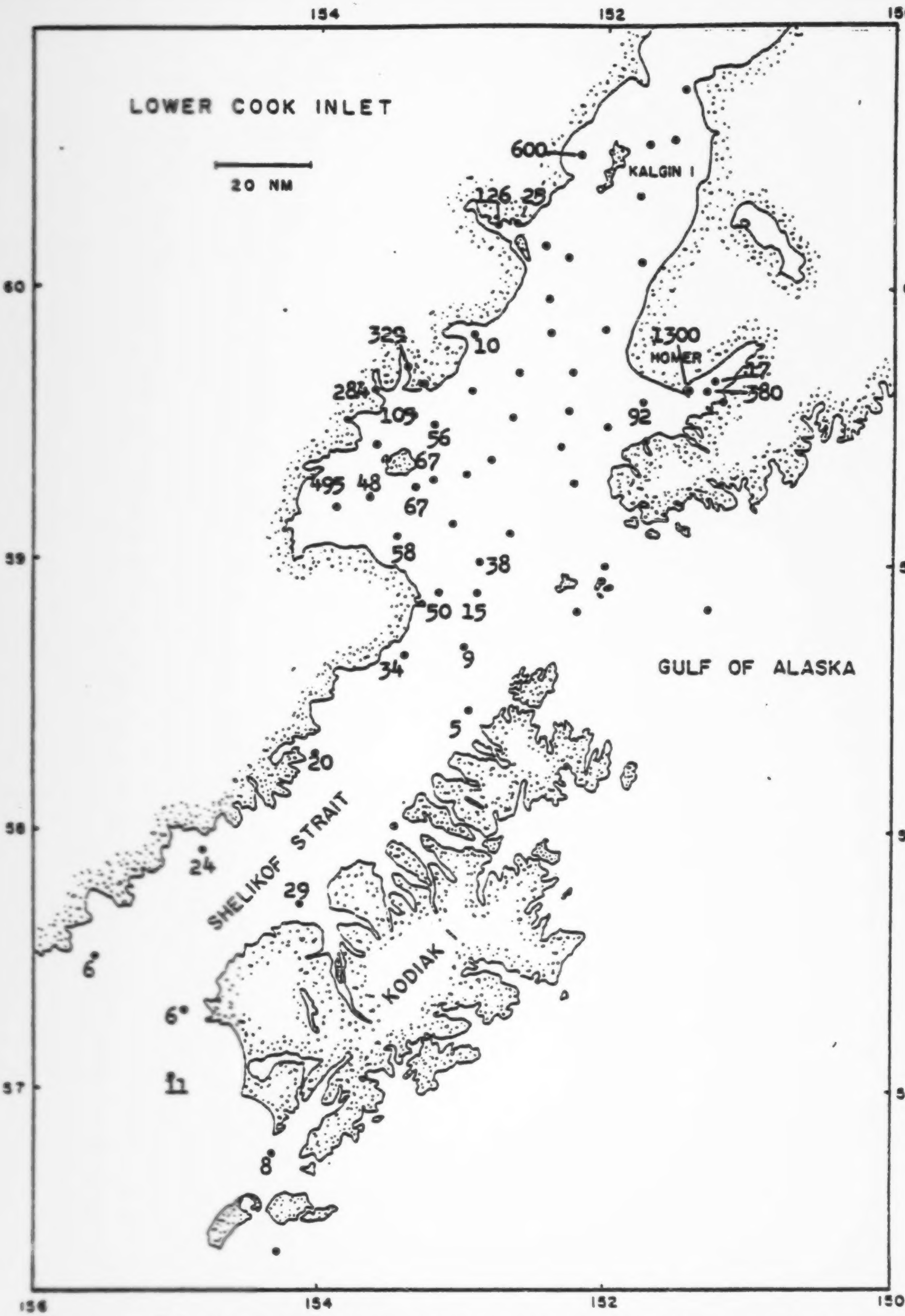

LOWER COOK INLET
20 NM
KALGIN I
HOMER
GULF OF ALASKA
SHELIKOF STRAIT
KODIAK I
600
126
25
329
10
1300
17
380
284
105
92
56
67
495
48
67
58
38
50
15
9
34
5
20
24
29
6
6
11
8
154
152
150
60
59
58
57
156

Figure 13. Relative microbial activity in the marine sediments collected during the April 1978 Cook Inlet cruise. Units used are ng glutamate/g/h.

all of the Cook Inlet cruises. There were differences observed in the mean rates; but when nitrogen fixation rates are compared on a station to station basis, there was no statistical significance in these differences (Table 4). During three of these cruises (11/77, 4/78, 4/79), there were enough samples taken in the Shelikof Strait so that a valid comparison could be made between sediment nitrogen fixation rates observed there and rates observed in the Cook Inlet (Table 4). In all three cases, the mean values observed in the Shelikof Strait were higher than those observed in the Cook Inlet. In two of these studies, the difference was significant at the $p<0.05$ level. The differences observed in Shelikof Strait nitrogen fixation rates between cruises were not statistically significant. The mean values for nitrogen fixation observed during both the Beaufort Sea cruises and the Norton Sound cruise were significantly lower than those observed in the Shelikof Strait (Table 4). In addition to the higher values observed in the Shelikof Strait, there were consistently high values observed in Kachemak Bay within Cook Inlet. These geographical trends are best illustrated in the November 1977 data shown in Figure 14.

C. Data storage

All of the cruise data is currently being stored at NIH under the direction of Dr. Krichevsky, RU #s 391 and 371. This file contains information on station location, salinity, temperature, cell concentrations, uptake rates with and without crude oil, respiration percentages with and without crude oil, nitrogen fixation rates with and without oil, and all inorganic nutrient data for

Table 4. Nitrogen fixation rates[1] in field station sediments 1977-1979.

		Apr. 1977	Nov. 1977	Apr. 1978	Apr. 1979	Sep. 1977	Aug. 1978	Jul. 1979
Cook Inlet[2]	$\bar{X}$	0.23	0.46	0.31	0.06			
	range	0-0.52	0.05-1.70	0.10-0.90	0-0.14			
Shelikof Strait	$\bar{X}$		1.3	0.51	0.68			
	range		0.3-4.4	0.30-1.10	0.45-0.95			
Beaufort Sea	$\bar{X}$					0.10	0.20	
	range					0.02-0.21	0-1.40	
Norton Sound	$\bar{X}$							0.22
	range							0-0.88

[1] rates expressed as ng N fixed/g dry weight sediment/hr.

[2] excluding Shelikof Strait and Homer boat basin.

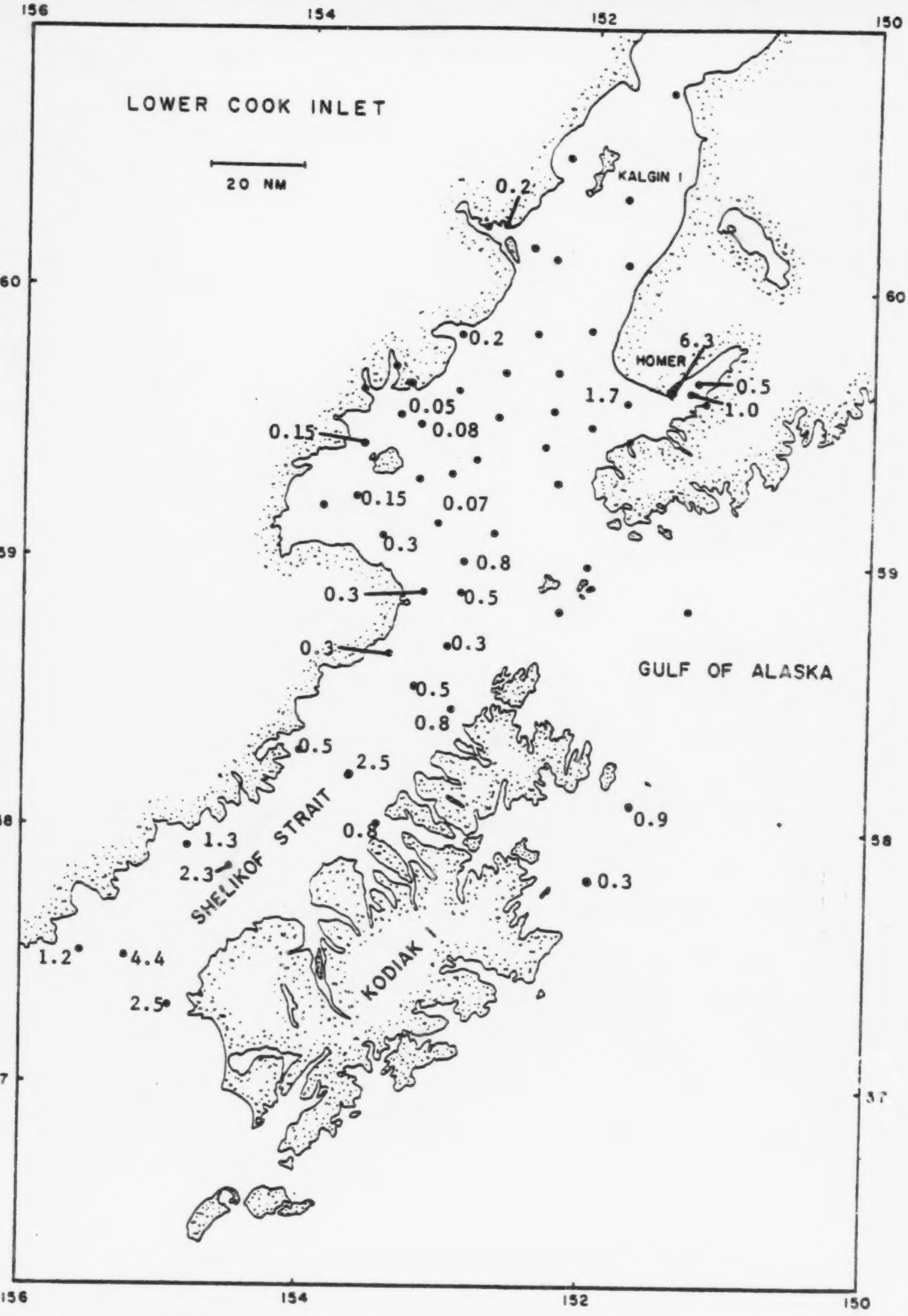

156
154
152
150
LOWER COOK INLET
20 NM
KALGIN I
HOMER
60
59
58
57
0.2
0.2
6.3
0.5
1.7
1.0
0.05
0.08
0.15
0.15
0.07
0.3
0.8
0.3
0.5
0.3
0.3
GULF OF ALASKA
0.5
0.8
0.5
2.5
0.9
0.8
1.3
SHELIKOF STRAIT
2.3
0.3
KODIAK I
1.2
4.4
2.5

Figure 14. Nitrogen fixation rates in sediment samples collected during the November 1977 cruise. The units are ng N_2 fixed/g dry wt. /h.

BEST DOCUMENT AVAILABLE

water and sediment samples. The data collected for each cruise are located in a different file. The file numbers for the Cook Inlet cruises are as follows:

	Sample # series	File #
October 1976	GW/B 300	237
April 1977	GW/B 400	254
November 1977	GW/B 500	259
April 1978	GW/B 600	306
April 1979	GW/B 800	260

V. Discussion

A. Relative microbial activity and percent respiration (mineralization).

1. Comparison between activities in sediments and the overlying water columns.

a. Our studies have shown that in regions where there are fine-grained sediments and the water column is relatively shallow, a vast majority of the microbial activity for the whole system resides in the sediments. As a result of our summer 1975 Beaufort Sea study, we concluded that in the shallow waters behind the barrier islands, where the average water depth at the sample locations was 3 meters, the relative microbial activity in the sediments was 400 times that found in the overlying column (see Section II). During the Beaufort 1975 study the ratio of the uptake rates in sediments/water was 14. This was computed by comparing uptake per liter of seawater with uptake per g dry wt of sediment. When the mean values for water and sediment uptake in the Cook Inlet are compared in the same way, the ratio is 16. Thus the relative importance of microbial activity in sediments compared to the water column is about the same in the Cook Inlet as that observed in the Beaufort Sea. Of course the mean water depth in Cook Inlet is greater than 3 meters. A more realistic figure would be 30 meters. Even at that depth, the microbial activity in the sediments should be at least 40 times greater than that in the overlying water column in areas where there are fine grained sediments.

b. Another difference that was observed between pelagic and benthic microbial function was the percent respiration values.

In all Cook inlet field studies, the mean values for both glucose and glutamic acid percent respiration in sediments were equal to or less than those observed in the water samples. The same phenomenon has been observed during our Beaufort Sea studies (see Section II). It is felt that this reflects a qualitative difference in the nutrients available to the microorganisms in these two environments. When the percent respiration is low, a greater proportion of the nutrients taken into the cells are being converted into new microbial biomass. This usually occurs when most of the essential growth factors are available to the organisms. This concept will be explained in greater detail later in this report.

c. The Cook Inlet studies have also shown that the level of suspended matter in the water column is also an important feature of the pelagic microbial environment. The comparison between relative microbial activity in offshore (10 meters from the surf zone) and surf zone waters have shown greater activity in the latter. This is undoubtedly due to the increased suspended matter load in the surf zone sediments with its associated microbial populations. A more direct connection between suspended particulates and relative microbial activity was observed in the waters of the Upper Cook Inlet. During two cruises, a comparison between relative microbial activity and water turbidity showed these two variables to be highly correlated (correlation coefficients of 0.87 and 0.89). It is quite possible that in high energy environments like those present in the Upper Cook Inlet, the most important locus of microbial activity may be in the water column instead of the sediments.

On the occasions that we have measured relative microbial activity in the sandy sediments of high tidal energy environments, we have found them to produce very low levels of microbial activity. This is opposite of what we have observed in low energy regions where fine grained sediments are found. The implications of these conclusions will be discussed below.

2. Seasonal differences

a. Unfortunately, all of the five Cook Inlet cruises in which we participated took place either in the spring during the month of April or in the late fall (October and November). Thus, the data generated from these cruises represent only two seasons making interpretation of seasonal variations very difficult (data was not available for the seasons of greatest extremes). In February 1979, we initiated a series of studies at a field station in Cook Inlet (Kasitsna Bay). Although our main purpose was to study the effects of crude oil on microbial processes, we did collect data on a large number of sediment and water samples at various times of the year. These data are reported in greater detail in Section IV. In summary, we found that microbial activity in the water column was highest in April after (or in association with) the spring diatom bloom and it was lowest in February. The microbial activity in the sediments showed much greater seasonal stability. The highest values were observed in August and the lowest in October and April.

b. Another interesting aspect of these studies was the fact that the ratio of glucose uptake to glutamic acid uptake changed seasonally. Glucose uptake was proportionately greater in pelagic populations during the time when there was greatest phytoplankton activity. This suggests that the microbial population was adjusting to the presence of organic nutrients released by the phytoplankton during periods of high primary productivity. Similar relationships have been suggested by Gillespie et al. (1976) and by Albright (1977).

c. With the data available from the Kasitsna Bay studies, it is possible to put the values of microbial activity which were observed during the Cook Inlet cruises in perspective. The glucose uptake rates and the ratios of glucose to glutamic acid uptake observed in the October, November and April 1977 cruises were taken under conditions that were closer to that expected under "winter" conditions. The April 1978 cruise was conducted at a time when the spring phytoplankton bloom was just beginning and the April 1979 cruise was conducted when the spring phytoplankton bloom was probably near its peak (at Kasitsna Bay, the diatom bloom had begun at least 4 weeks before the cruise began). During this cruise, we observed microbial activity patterns which did not fit those that had been observed during any other cruise. There were spikes of very high activity throughout the Cook Inlet and Shelikof Strait. This indicated that there was a great deal of patchiness in the occurrence of these blooms. This is consistent with the data reported by Dr. Larrance in his final report for RU #425 (Annual Reports of Principal Investigators for the Year ending March, 1977. Vol 10:1-136).

3. Geographical differences

a. As was mentioned in the Results section, consistent trends in surface water relative microbial activity and percent respiration patterns were observed. The resulting patterns, when interpreted in light of what is known about the hydrography and chemistry of the region, produce an overall picture of the dynamics of the system which will assist those in government and industry in making a more accurate assessment of the potential problems related to crude oil production in Cook Inlet.

A discussion of the conclusions drawn from these data as well as the facts and assumptions on which these were based have already been mentioned in section I.B of Section I. An analysis of these data have also been submitted for publication (Griffiths et al., 1980d). The following is an amplification of the data presented in that section.

There are two distinct water masses present in Cook Inlet; one to the north that is very turbid and of relatively low salinity and one to the south and southeast which is more typical of open ocean water. We have found that both of these water masses have characteristic patterns of microbial activity and respiration. Glutamic acid uptake studies in surface waters have shown that the relative microbial activity is very high and the respiration percentages are very low in the northern water mass. The reverse pattern is seen in the water mass to the south. Intermediate values were observed in regions where these two water masses meet in the area to the north and east of Augustine Island. This is the same region in which a gyre has been observed by other investigators. In general, the patterns of surface water microbial activity and respiration reflect the net surface circulation patterns reported by Miller and Allen (1976).

As far as we know, this is the first study made in which patterns of microbial activity in marine waters have been used to characterize more than one distinct water mass and to indicate regions of interaction between those water masses. The two water masses in question are clearly shown by the surface water salinity data illustrated in Fig. 9. This is a similar pattern to that observed by Kinney at al. (1969). This is also the same type of pattern that one would expect from the current data presented by Miller and Allen (1976). Since these observations were taken at various times, it would appear that this is a relatively consistent feature in Cook Inlet. These same patterns are clearly shown in the relative levels of microbial activity and respiration percentages observed in the same region (Figs. 10 and 11) during all three Cook Inlet cruises.

At this point it is important to reflect on what these observations mean in terms of what is occurring in these water masses. The water mass to the southeast is coastal water which is being pushed into the inlet by inshore currents moving to the west. These waters probably contain very low levels of available organic nutrients. As a result, the level of microbial activity is low and the percent respiration is high. It has already been established by a number of investigators (Wright and Hobbie, 1966; Vaccaro and Jannasch, 1966; Crawford et al., 1974; and Carney and Colwell, 1975) that the uptake rate of simple labeled amino acids and sugars by natural microbial populations usually reflect the levels of nutrients present in the surrounding water. The significance of the percent respiration data is less clear. A relatively high percent respiration value indicates that the population is using proportionately more of the nutrient as an energy source and less of it to produce cellular material. There are at least two conditions in which this might occur. If the cells are starved, the cells will utilize most of added nutrient for energy requirements before biosynthesis is initiated. A more likely explaination is that growth factors (available nitrogen or phosphorus) are not present in sufficient concentration to allow biosynthesis to occur even though nutrients are available to the cells.

The high levels of relative microbial activity and low respiration rates found in the northern waters indicate that these waters contain nutrients that are qualitatively and/or quantitatively different than those found in the southern waters. The regions where these two water masses mix show intermediate values between these two extremes. These intermediate values could be caused by at least two factors. As the northern water mass moves south along the measured edge of the inlet, the nutrients present are being consumed by the microorganisms present. At the same time, low nutrient waters from the south are being mixed with other water thus diluting the nutrients.

Drs. Cline and Feely (1977) have shown in their studies that the water masses in Cook Inlet are usually well mixed vertically. We measured relative microbial activity at three locations and at various depths during the November cruise. We found no significant microbial activity stratification with depth. It would thus appear that the observations made in the surface waters should hold true for the entire water column in most locations.

We have also observed that the relative levels of microbial activity are directly related to the levels of suspended matter in the surface waters. When relative microbial activity as measured using glutamic acid is compared with turbidity in the same samples, correlation coefficients of 0.87 and 0.89 were observed for all water samples collected during the April and November 1977 cruises respectively. The correlation is also substantiated by the suspended matter patterns reported by Feely and Cline (1977). There is a striking similarity between these patterns and the patterns of microbial activity and respiration percentages reported here. During our determinations of bacterial concentrations using epifluorescent microscopy, we have observed that 70-80% of the bacteria present in water samples are associated with the particulate matter.

Feely and Cline (1977) also reported that much of the suspended matter found in the northern waters probably makes its way into the sediments of the Shelikof Strait. Our studies of relative microbial activity in the Cook Inlet tend to support this hypothesis. During all cruises, relatively high rates of microbial activity were observed in the sediments of Tuxedni Bay, Kachemak Bay and the southern portion of Kamishak Bay. The high rates of activity seen in Kachemak Bay are probably due to the trapping of nutrients within the bay. It has been observed by other investigators that the net flow of water through this bay is very low.

The high levels of microbial activity observed in Tuxedni Bay sediments are probably due to the sedimentation of the microbiologically active suspended matter in the water column in this area. The high levels of microbial activity in the sediments of the southern Kamishak Bay area could be the result of organic nutrients being transported from either the Upper Cook Inlet as described above or from the sedimentation of planktonic material from Lower Cook Inlet.

b. The consistently high rates of microbial activity found in Kachemak Bay may have a direct bearing on the high fisheries productivity in the same region. Dr. Feder (RU #281) has conducted studies of the food webb in Cook Inlet. He has reported that both littoral and offshore benthic organisms are very much dependent on detrital food sources (Lower Cook Inlet, Alaska - A Preliminary Environmental Synthesis). He has also reported that immature crab and adult shrimp ingest sediment directly to extract the nutrients contained in the sediments. From what is now known about detrital food chains, it can be assumed that these organisms are obtaining most of their nutrients from the bacteria that are associated with the detrital particles within the sediment. D. C. Lees (RU #417) has also reported (Final Report - Reconnaissance of the Intertidal and Shallow Subtidal Biotic - Lower Cook Inlet, 1977) that the productivity of macrophytes is higher in Kachemak Bay than in any other region in Cook Inlet. The growth rates found here were as high as any reported in the current literature. Recent studies of macrophyte decomposition and utilization of macrophyte biomass by other organisms indicate that ca. 80% of this biomass is routed through bacteria before it can be used as a usable food source by the higher trophic levels (Fenchel and Jorgensen, 1977). There is therefore, a great deal of presumptive evidence that benthic bacteria play a vital role in the overall productivity of this region.

B. Nitrogen fixation

1. Seasonal differences

a. Fixed nitrogen is one of the limiting nutrients in the ocean and therefore productivity of any marine ecosystem relies on the availability of nitrogenous compounds. The only mechanism that can fix N_2 in the marine environment is enzymatic reduction by certain microbes (including the cyanobacteria). As was the case in interpreting the relative microbial activity on a seasonal basis; it is very difficult to make statements about seasonal nitrogen fixation based on the results of the cruises. Fortunately, during the Kasitsna Bay study, we conducted measurements of nitrogen fixation on a seasonal basis (see section IV). These results show that the highest rates of nitrogen fixation occurred in the months of February, April and October and that the lowest rates occurred in July 1979. On a seasonal basis, April nitrogen fixation rates in the Beaufort Sea were the highest observed in that region. Although the differences observed in the mean values for nitrogen fixation during the Cook Inlet cruises were not statistically significant, it is interesting that the November 1977 rates were the highest recorded in both Cook Inlet and Shelikof Strait.

The reason for these seasonal variations is probably related to both the quality and quantity of the nutrients coming into the sediments. The stimulation of nitrogen fixation by the addition of soluble organic carbon has been well documented in the literature (Knowles and Wishart, 1977; Fay, 1976; Keirut Brezonik, 1971; and Herbert, 1975).

2. Geographical differences

During the cruises, we consistently found the highest rates of nitrogen fixation in Kachemak Bay and Shelikof Strait. Occasionally, high nitrogen fixation rates were also observed in Kamishak Bay as well. These patterns can be interpreted in light of data collected at Kasitsna Bay, data collected on the currents in Cook Inlet and the denitrification data collected by Dr. Atlas and his associates. The current patterns described by Miller and Allen (1976) suggest that the net flow of water out of the Inlet is south along the west side of the inlet and Shelikof Strait. This pattern is consistent with the results of studies conducted by Feely and Cline (1977) and ourselves. It can thus be assumed that detrital particles that are produced in Cook Inlet, would move to the west side of the Inlet and then south through Shelikof Strait.

It seems likely that as the particles migrate south, the ratio of carbon:nitrogen (C:N) increases as the bacteria use up the available nitrogen and are cropped (a similar mechanism is suggested for the seasonal changes observed in the Kasitsna Bay study). Particles that settle out in Kamishak Bay would have lower C:N ratios than those found in Shelikof Strait which is further to the south. Qualitatively, the organic nutrients found these sediments would then be similar to those found in the "winter" sediments of Kasitsna Bay. This would explain why the relative microbial activity is low and the nitrogen fixation rates are high in the Shelikof Strait and the reverse pattern is found in Kamishak Bay.

Additional support for this hypothesis comes from a comparison of nitrogen fixation and denitrification rates in these two areas

(Table 5). These data are also presented by Haines et al. (1980). In the western Cook Inlet (Kamishak Bay), the natural rates of denitrification (the conversion of fixed nitrogen to atmospheric nitrogen) were much greater than natural rates of nitrogen fixation (the opposite process). These rates are probably out of balance because of an exogenous input of fixed nitrogen into this area; i.e. detrital particles with low C:N ratios. This is the same situation that was observed in Norton Sound where the exogenous source of fixed nitrogen was terrestrial detritus from the Yukon River. In the Shelikof Strait, the situation was different. The amount of nitrogen being fixed was approximately equal to the amount of fixed nitrogen that was being transformed back to atmospheric nitrogen. This would suggest that there was very little exogenous nitrogen being supplied to the system.

3. Significance of nitrogen fixation in overall productivity

The nitrogen fixation rates observed in Shelikof Strait and Kachemak Bay were significantly higher than those rates observed in either the Beaufort Sea or Norton Sound. This indicates that this process is probably of greater significance in and near Cook Inlet than in Arctic waters. How then do these rates compare with those reported by other investigators in different areas? Of the studies reported, the one that most closely approximates ours was that of Herbert (1975). This was an *in situ* study of nitrogen fixation in sediment cores taken at a location on the northeast coast of Scotland. Herbert observed a maximum nitrogen fixation rate of 1.84 ng nitrogen fixed per g dry wt per h. This rate is the *average* rate of nitrogen fixation observed by us in all sediment samples analyzed during the November 1977 Cook Inlet cruise. The maximum rate that we observed was 6.3 ngN x g^{-1} x h^{-1}. In another study, Brooks et al. (1971) reported a range of nitrogen fixation in 8 sediments taken from a Florida estuary of from 0.64 to 6.0 ngN x g^{-1} x h^{-1}. The highest value that they observed was very close to the highest value that we observed in November Homer boat basin). Marsho et al. (1975) reported an average annual nitrogen fixation rate of 2.9 ngN x g^{-1} x h^{-1} in sediments taken from 7 stations in the Rhode River close to Chesapeake Bay. These data suggest that the rates of nitrogen fixation that we observed in Cook Inlet and the Shelikof Strait are close to that observed in other marine sediments and relatively high when compared to sediments that were most similar (the Herbert study).

Table 5. Comparison of mean rates of nitrogen fixation, denitrification and concentrations of fixed forms of organic nitrogen in sediments from different regions of the Alaskan Continental Shelf.

	N_2 fixation ng g^{-1} h^{-1}	Denitrification (natural) ng g^{-1} h^{-1}	NH_4^+ µM	$NO_3^- + NO_2^-$ µM
Upper Cook	0.08	0.05	-	-
Western Cook	0.28	11.25	137	55.2
Shelikof Strait	0.68	0.66	83	3.1
Norton Sound	0.22	1.91	188	6.4
Beaufort Sea	0.20	-	103	12.4

In the Kasitsna Bay study, we found that the yearly mean value for nitrogen fixation rates in sediments analyzed near there as being approximately 1 ng x g dry wt^{-1} x h^{-1}. This rate was sufficient to replace all of the fixed nitrogen (NH_4^+, NO_3^-, and NO_2^-) in the interstitial water every 24 hours. It is also a rate that is sufficient to account for an annual production of bacterial biomass in Kachemak Bay of 400 tons.

Under most conditions, nitrogen is usually not limiting to bacterial growth in seawater; however, the same may not be true in sediments. Inshore sediments often contain detritus particles which have very high carbon to nitrogen ratios. Studies of detrital food chains have shown that nitrogen fixation in sediments may be a very important factor in the effective utilization of detritus food particles by higher trophic levels (Mann, 1972; Fenchel and Jørgensen, 1977). This is particularly important when one realizes that the majority of organic nutrients available to support all of the animal population in the inlet probably come from detritus particles. In order for this to become available as a food source for animals from the level of the protozoa on up, the detritus particles must become colonized by bacteria. In order for bacteria to grow, they need fixed nitrogen.

IV. Conclusions

1. In general, the sediments showed very high microbial activity when compared to the water samples studied. This fact suggests that the sediments might be important loci of crude oil degradation. It is not known what rates of biodegradation could be expected in Arctic marine sediments; however, there is evidence that suggests that certain components

of crude oil may persist in sediments for long periods of time (see Atlas' final report for RU#29).

2. Water samples taken along the shoreline consistently show higher levels of microbial heterotrophic activity than those taken offshore. This indicates that initial rates of crude oil biodegradation may be higher in waters next to the beach than in offshore waters.

3. The waters taken from the northern section of Cook Inlet showed much higher levels of activity than those taken near the mouth of the inlet.

4. Measurements of relative microbial activity and respiration percentages can be used to characterize specific water masses and give some information about the organic nutrients found in these waters. The above measurements should be made in both the water column and sediments in new lease areas where this information is not available. These data would provide information concerning the microbial characteristics of water masses as well as data concerning biological productivity potential.

5. Nitrogen fixation in the sediments of Cook Inlet and the Shelikof Strait may be an important contributing factor to the overall productivity of the detritus based food chain in that area.

6. Our studies on the effects of crude oil on nitrogen fixation in natural sediment samples showed that the presence of crude oil had little or no short-term (24 h or less) adverse effect on this process. As indicated in Section IV, long-term exposure of marine sediments did cause a dramatic decrease in nitrogen fixation rates.

7. Crude oil did have an inhibitory effect on glucose respiration in natural marine microbial populations. This effect was noted when either

crude oil, crude oil aqueous extract or weathered crude oil was used. This effect probably reflects stress which could cause a reduction in species diversity such as that already observed in Arctic marine waters exposed to crude oil over extended periods.

8. Evidence is accumulating which suggests that crude oil which is spilled in the turbid waters of the Upper Cook Inlet may become associated with the suspended matter found in these waters. If this occurs, then crude oil components would become associated with the sediment when these particles settle out of the water column. Our studies and the studies of Drs. Feely and Cline suggest that these particles probably settle out into the sediments of the southern Kamishak Bay and/or the sediments of the Shelikof Strait.

9. The regions which would most probably be impacted by a crude oil spill would be Kachemak and Kamishak Bays and the Shelikof Strait. This is assuming that the crude oil becomes associated with the sediments. Of these three areas, the one which would be most severely impacted would be Kachemak Bay. If significant portions of the sediment in this Bay was perturbed with crude oil, it is quite likely that the fisheries in this region would be adversely effected for an extended period of time because of the impact on the detrital food chain.

10. Our effects studies at Kasitsna Bay have shown that virually every benthic microbial process that we have studied show significant changes when marine sediments are exposed to crude oil. These results are reported in Section IV of this report.

VII. Needs for further study

We have collected all of the field data in Cook Inlet and Shelikof Strait required to have a general understanding of relative microbial activity and nitrogen fixation rates in these areas. Future cruise work should be conducted in the Bering Sea where there is very little information available about microbial processes. We do, however, strongly recommend that the effects studies that we have initiated at Kasitsna Bay be continued.

BEAUFORT SEA

Section II

I. Summary of objective, conclusions and implications with respect to OCS oil and gas development.

A. Objectives.

During the early studies, our main objectives were to obtain baseline data on microbial activity and bacterial cell concentrations in the waters and sediments of the Beaufort Sea. These data were to be augmented with salinity, temperature and inorganic nutrient data. These studies were to be designed to define when and where the microbial populations were the most active. As the study progressed, we were to start measuring nitrogen fixation rates in the sediments and the short-term effects of crude oil on these processes. In July 1977, a study was initiated in Elson Lagoon which was designed to show what effects, if any, crude oil had on benthic microbial populations. These studies have been conducted from that time to the present. During all of these studies, we were to work in close cooperation with Dr. Atlas (RU #29).

B. Conclusions and implications with respect to OCS oil and gas development.

1. Our studies to date have shown that crude oil has significant long and short-effects on microbial function in Arctic and sub-Arctic marine environments. This has some important implications concerning the overall productivity of an impacted area. Some of the long-term effects of crude oil in sediments include reduced rates of organic nutrient mineralization. This and reduced trans-

formation rates of key nitrogen cycle components suggest that crude oil could reduce the rate at which inorganic nutrients such as nitrate and phosphate are made available to form new phytoplankton biomass used in primary productivity.

2. There is also evidence to suggest that secondary productivity, through the detrital food chain, could also be adversely affected by the presence of crude oil in sediments. Such a reduction in secondary productivity would have a profound effect on the food available to support life at all trophic levels. This reduction would most probably be of greatest importance in the shallow inshore communities; especially those areas near the major rivers from which terrestrial carbon is transported seasonally. Recent studies on the dynamics of detrital food chains suggest that over 50% of phytoplankton carbon and 80-90% of macrophytic and terrestrial carbon passes through bacterial biomass before being utilized by higher trophic levels. We have typically observed reductions in microbial activity of 50% or more in waters and sediments exposed to crude oil. By inference, crude oil should have a profound effect on overall biological productivity in an impacted area. The aquaria studies at Kasitsna Bay have shown that there is a large accumulation of detrital material on the surface of oiled sediments. This did not occur in the non-oiled control sediments (see Section IV for details of this study). Under these conditions, the blockage of the detrital food chain by the presence of crude oil was dramatically illustrated.

3. Beaufort Sea inshore sediments were shown to have approximately 400 times the microbial activity as that found in the overlying

water column. This illustrates the relative importance of the sediments in the system. As mentioned above, it is known that crude oil has a profound effect on microbial activity in these sediments. We therefore recommend that during the planning and execution of oil production, those procedures which could cause crude oil to become accidentally incorporated into the sediments should be avoided. Likewise, procedures which would tend to drive crude oil into the sediments during cleanup operations after a spill should be avoided.

4. The onset of measurable long-term crude oil effects took considerably longer in Beaufort Sea sediments than they did in Cook Inlet sediments (up to 10 times longer). It seems quite likely that the crude oil effects that we are observing in the Elson Lagoon samples will persist for a long time. We have seen many of these effects persisting in Cook Inlet sediments one year after exposure. From these observations and what we know about what takes place in Beaufort Sea sediments, we would predict that crude oil would affect benthic microbial function for several years.

5. In a recent study conducted by Dr. Schell (RU #537), it was concluded that the Beaufort Sea amphipod Gammarus steosus feeds directly on peat. This shows a direct link between cellulose utilization and transfer of this carbon to higher trophic levels. Actually, the amphipods are probably digesting off the bacteria which colonize the peat. It has also been shown by Busdosh and Atlas (1977) that Beaufort Sea amphipods avoid feeding in areas contaminated with crude oil. In our Kasitsna Bay study, we also

observed a reduction in the utilization of detritus because the detritivores were either killed or avoided feeding in crude oil contaminated sediments. This leaves us with the question of what happens to the detritus if it is not utilized by the detrital food chain? Our work at Kasitsna Bay shows that the detritus accumulates on the surface of the sediments and the sediments become anaerobic. These anaerobic conditions would undoubtedly effect the long-term distribution of the normal benthic organisms since most of the sediments we have sampled in the Beaufort Sea appear to be well oxidized.

II. Study areas.

During the summer of 1975, we conducted a two month study of microbial activity in marine waters and sediments near Point Barrow and Prudhoe Bay. The locations that were sampled are illustrated in Figs. 15 and 16. At this time, 50 water samples (including 5 ice samples) and 24 sediment samples were collected and analyzed. During the 1976 field studies the locations shown on Fig. 17 were sampled. During April, 1976, 26 water samples (including 3 ice samples) and 14 sediment samples were analyzed. During the August 1976 Glacier cruise 18 water and 13 were collected. The sediment samples that were returned to us from the September 1977 cruise were sampled at the locations given in Fig. 18. During the August 1978 cruise we collected and analyzed 42 water and 38 sediment samples from the locations given in Fig. 19. The location for the Elson Lagoon oiled tray experiment was approximately 1/2 way between stations 2 and 3 in Fig. 16.

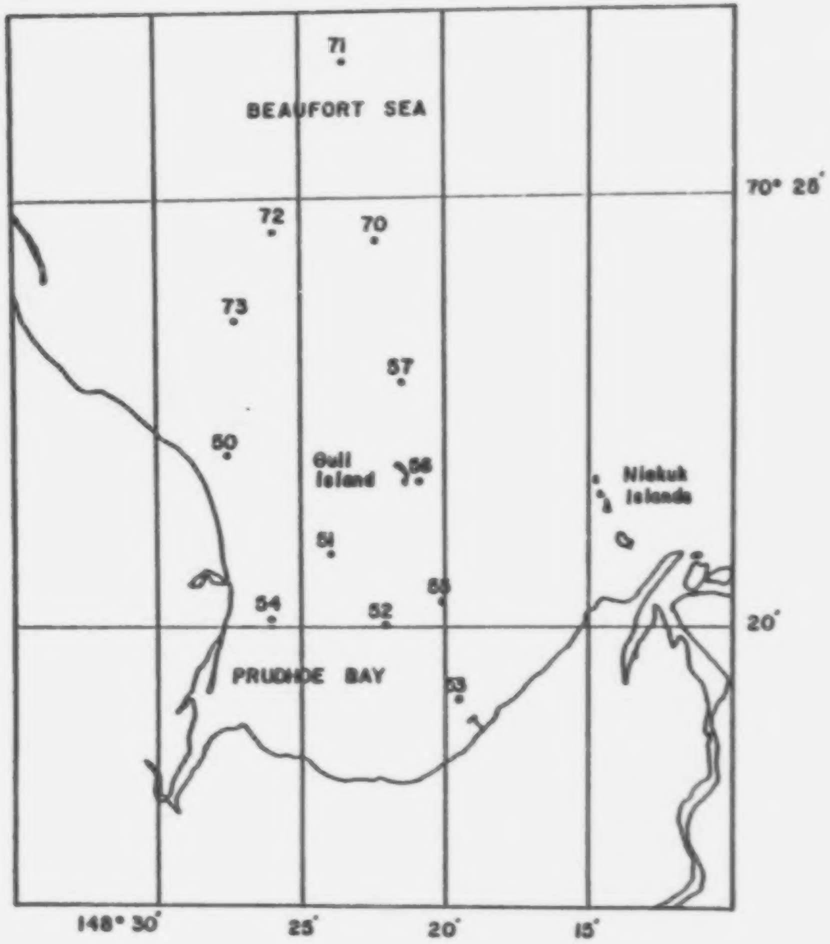

71
BEAUFORT SEA
70° 28'
72
70
73
57
50
Gull Island
56
Niakuk Islands
51
55
54
52
20'
PRUDHOE BAY
53
148° 30'
25'
20'
15'

Figure 15. Stations sampled in the Prudhoe Bay area during April 1976.

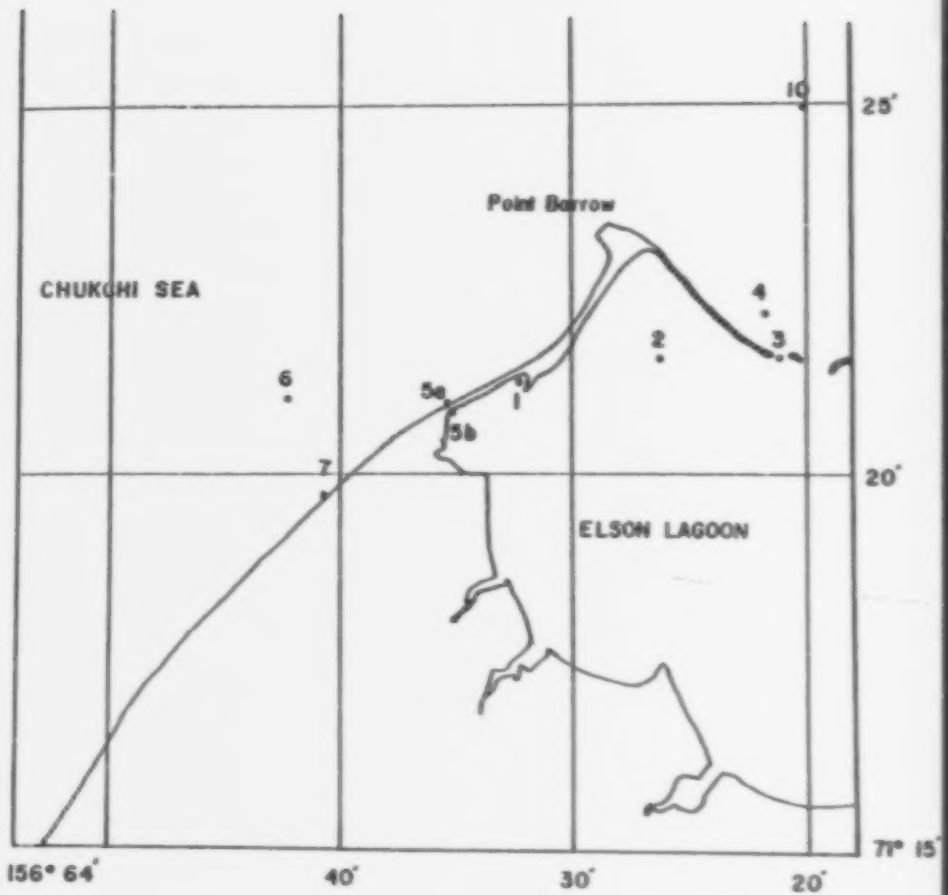

10
25'
Point Barrow
CHUKCHI SEA
4
2
3
6
5a
1
5b
7
20'
ELSON LAGOON
71° 15'
156° 64'
40'
30'
20'

Figure 16. Stations sampled in the Barrow area during April 1976.

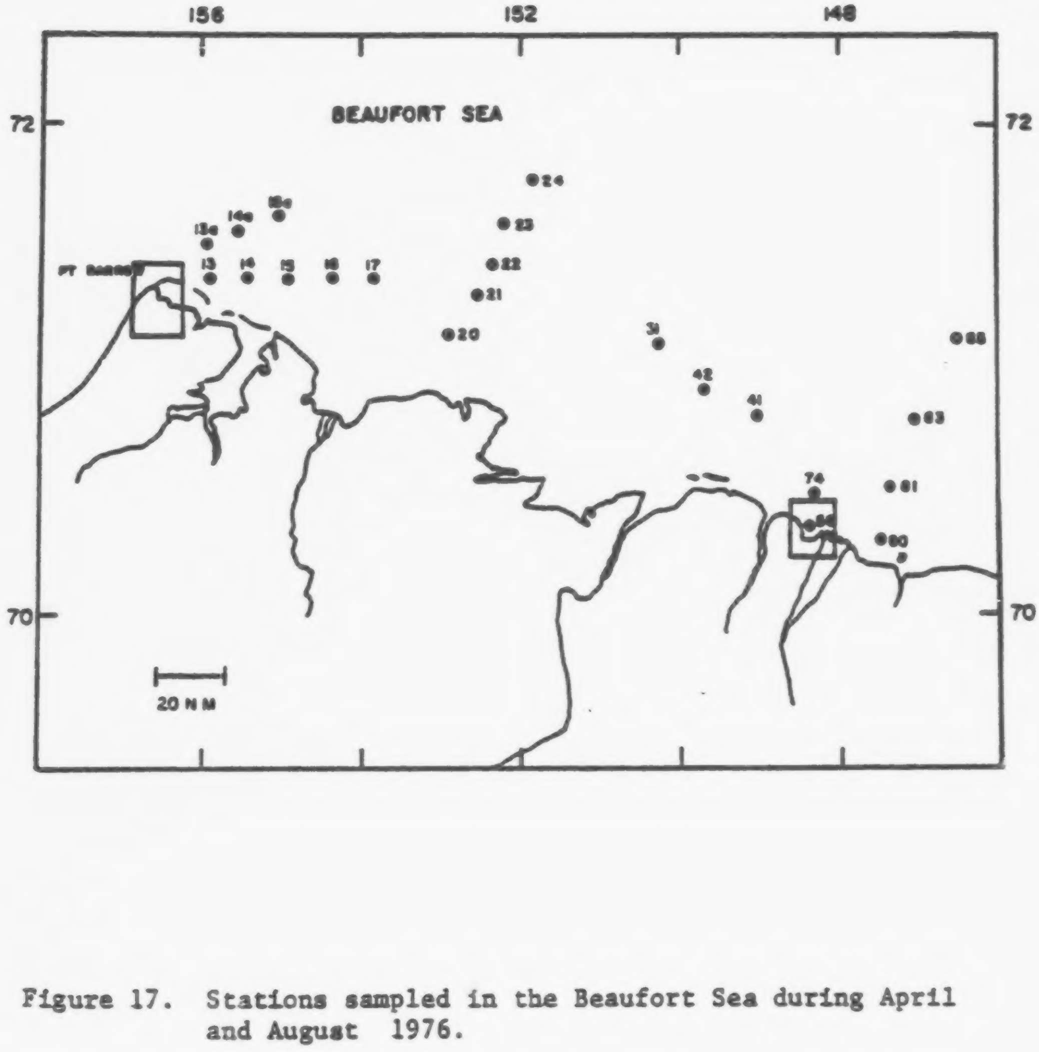

156
152
148
72
72
BEAUFORT SEA
24
23
22
21
20
15a
14a
13a
PT BARROW
13
14
15
16
17
31
42
41
85
83
81
74
80
70
70
20 N M

Figure 17. Stations sampled in the Beaufort Sea during April and August 1976.

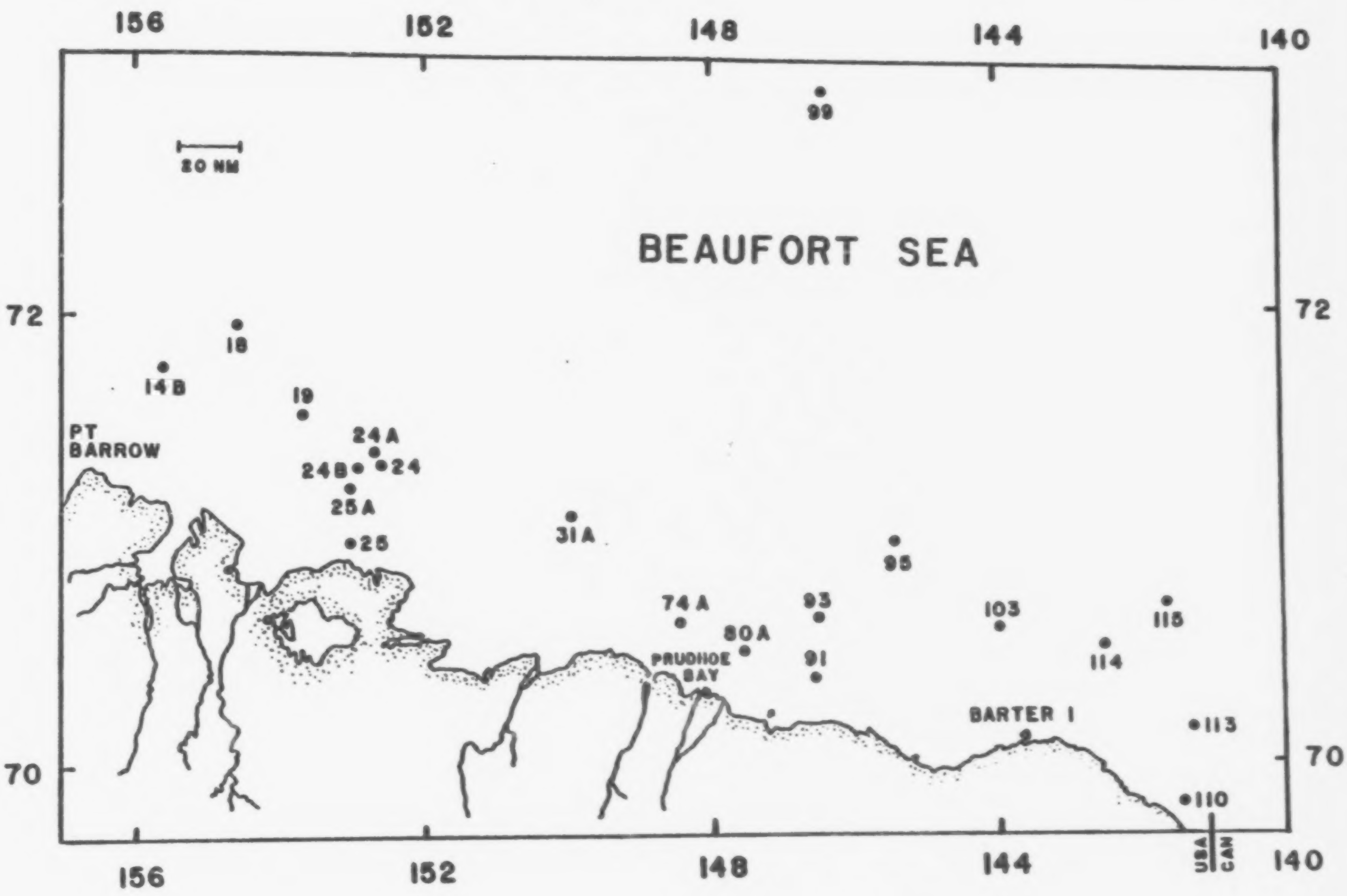

156
152
148
144
140
72
70
20 NM
BEAUFORT SEA
PT BARROW
14 B
18
19
24 A
24 B
24
25 A
25
31 A
74 A
80 A
PRUDHOE BAY
93
91
99
95
103
114
115
113
110
BARTER I
USA
CAN

Figure 18. Stations sampled in the Beaufort Sea during the September 1977 cruise.

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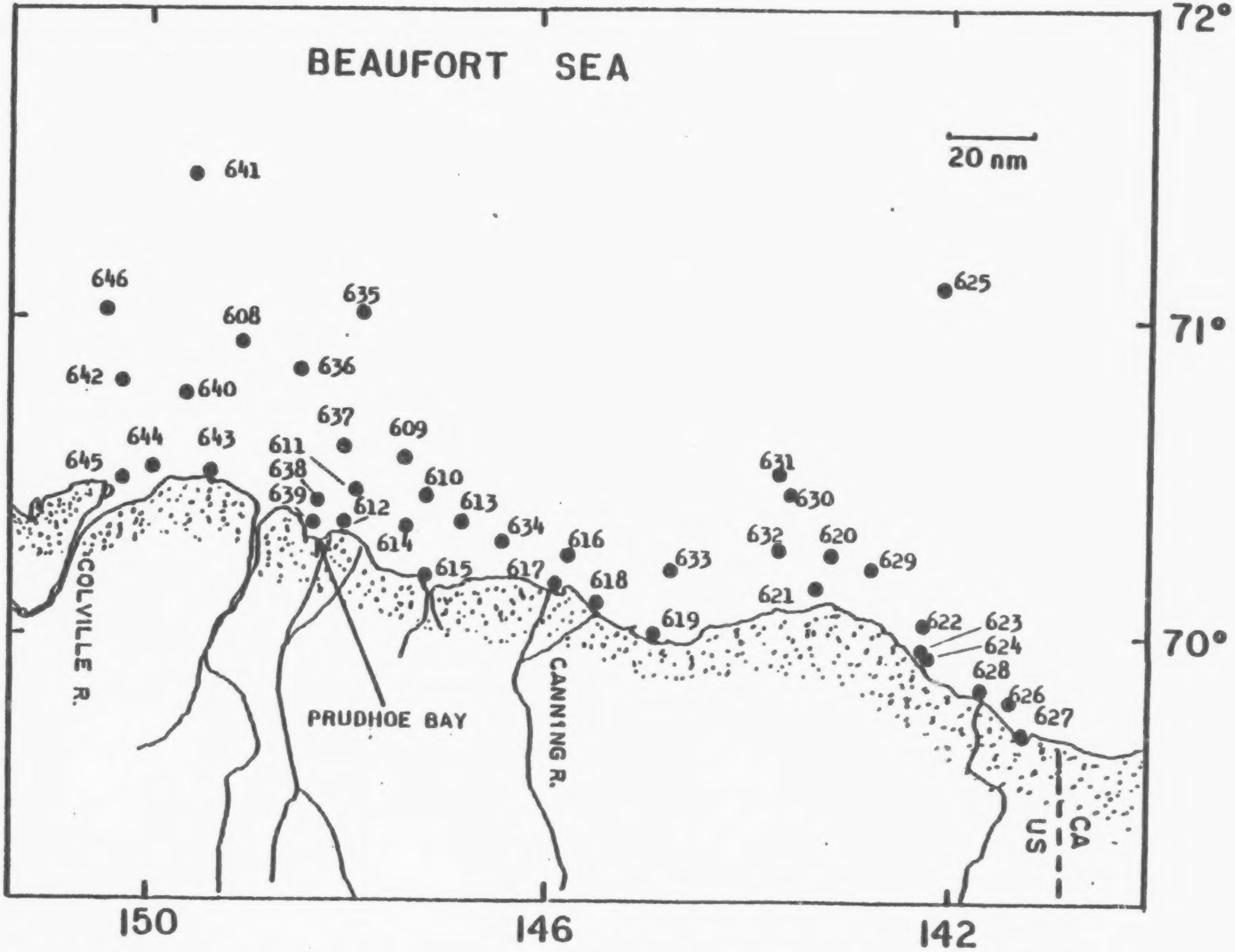

BEAUFORT SEA
20 nm
72°
71°
70°
150
146
142
641
646
635
625
608
636
642
640
637
609
644
643
611
645
638
610
639
612
613
614
634
616
615
617
618
633
631
630
632
620
629
621
619
622
623
624
628
626
627
COLVILLE R.
PRUDHOE BAY
CANNING R.
US
CA

Figure 19. Stations sampled in the Beaufort Sea during the August 1978 cruise.

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III. Methods.

The methods used for most routine field operations are presented in Section I. The methods used in the crude oil and Corexit effects studies are presented in Section IV of this report. The only method that is not covered in the other methods sections in this report was the one used to measure ATP, ADP, and AMP.

A. Analysis of adenylate concentrations

The procedure was a modification of that described by Bulleid (1978). One ml of sediment was added to 8 ml of extraction buffer (0.04 M Na_2HPO_4 adjusted to pH 7.70 with 0.02 M citric acid) in a 50 ml beaker which was placed in a boiling water bath for 2 minutes. With the exception of the time when the samples were boiled they were kept cold by placing the beakers in crushed ice. Evaporation was minimized by covering the beakers with a watch glass. To determine the extraction efficiency, 0.1 ml of 10^{-6} M ATP was added to both one sediment sample and the control containing no sediment. Duplicate subsamples of each sediment were extracted. Following extraction, samples were removed to an ice bath, then added to centrifuge tubes, allowing a small volume distilled H_2O rinse to completely recover the extracted sample. After centrifuging at 800 x g at 0°C for 10 min., the clear supernatant was removed to a screw cap vial, its volume brought up to 10.0 ml with distilled H_2O, and frozed at -20°C until the time of analysis.

For the analysis of ATP, 1.8 ml of extract was added to 0.72 ml of buffer 1 (0.55 g K_2SO_4 and 1.5 g $MgSO_4$ in 10.0 ml distilled H_2O) and 0.2 ml Tris-EDTA buffer (buffer A: 20 mM Tris; 2 mM EDTA; pH 7.75).

For analysis of ATP plus ADP, 1.8 ml of extract was added to 0.72 ml of buffer B (10 ml of buffer A with 5 mg phosphoenol pyruvate) and 0.2 ml pyruvate kinase (Sigma) solution (10 mg in 10 ml TRIS-EDTA). For analysis of ATP plus AMP, 1.8 ml of extract was added to 0.72 ml buffer B, 0.2 ml pyruvate kinase, and 0.05 ml adenylate kinase solution (Sigma). Each mixture was held on ice during preparation, then incubated at 30°C for 15 min and returned to an ice bath.

An Aminco ATP Photometer and Integrator-timer was used for the assay. a 575 µl volume of each mixture was transferred to a cuvette and placed in the photometer chamber. Integration of photon emissions over 3 sec intervals was begun as soon as 100 µl of a buffered luciferin-luciferase preparation (Dupont Instruments, No. 750145-902) was injected into the chamber through a septum. The maximum integrated count, usually occurring in the second interval, was used to calculate amounts of each adenylate present, using standard curves prepared from pure adenylate solution.

The concentrations of adenylates in each sample were calculated using a standard curve which was prepared by assaying known concentrations in the approximate concentration range expected in the samples. Once the concentrations in a sample were determined, they were normalized to grams sediment dry weight. An extraction efficiency was calculated for each sample which was then used to calculate the concentration of adenylates in the original sample. The energy charge was calculated using the following relationship:

$$\text{Energy Charge} = \frac{\text{ATP} + 1/2\ \text{ADP}}{\text{ATP} + \text{ADP} + \text{AMP}}$$

IV. Results.

Our first study in the Beaufort Sea was conducted during the months of July, August and September 1975. This study resulted in an extensive survey of relative microbial activity in the waters and sediments near Barrow, Prudhoe Bay and landward of the Barrier Islands between these two locations (Figs. 15 and 16). These first studies were published in 1978 (Griffiths et al.). We also participated in two cruises in the Beaufort Sea. During the first cruise (August 1976 Glacier cruise), we studied relative microbial activity at offshore locations between Point Barrow and Prudhoe Bay (Fig. 17). During our second cruise conducted in August 1978, we studied relative microbial activity and nitrogen fixation rates in samples collected from the Colville River to the US-Canadian border (Fig. 19). We did not participate in the September 1977 Glacier cruise, but Dr. A. G. Carey (OSU) brought back sediment samples to us. We measured nitrogen fixation rates and hydrocarbon biodegradation potentials in these samples (Fig. 18).

We have also participated in another study in collaboration with Dr. Atlas (RU #129). This study was designed to assess the effects of crude oil on microbial function in Elson Lagoon sediments. The results of these studies are presented and analyzed in Section IV of this report.

A. Relative microbial activity and respiration percentages.

In the studies mentioned below, the relative microbial activities are expressed in V_{max} values. This is equivalent to the substrate uptake rates used in the previous section.

1. Local variations.

Variations in the maximum potential uptake rates (V_{max} values) of water samples taken at the same location within a few weeks of one another are shown in Table 6. In some cases, the variation seen in the water samples was greater than 10-fold at the same station. The variation was somewhat less in the sediment samples. These variations are much greater than one would expect from experimental error alone since the variation between identical subsamples is generally near 10% for the technique used. The variations seen in the water samples probably reflect differences in water masses present at each location. This supposition is supported by the fact that stations sampled on the same date usually showed the same relative pattern of microbial activity even though the average level of activity may change dramatically from one week to the next. All of these samples were taken near or within the Barrier Islands in very shallow seas. These waters are greatly influenced by wind-driven currents and the effects of freshwater runoff. The variations in the sediments probably reflect inaccuracies in station location and the patchiness of the sediment due to ice gouging.

2. Geographical differences.

During the first three field-study periods, bacterial concentrations, relative microbial activity (V_{max}), temperature, salinity, and pH were measured in water and/or sediment samples. During the summer 1975 study period, these measurements were compared in samples that were taken in the Barrow and Prudhoe Bay areas (Table 7). In the water samples, there was a significant difference seen between the mean percentage of respiration and the mean salinity values observed in these two regions. Both values were greater in the Barrow area.

Table 6. Variations seen in V_{max} values observed in samples taken at the same geographical location at different times.

Station No. summer 1975	Water samples	
	Date	V_{max}^{*}
1	9/5	51
1	9/11	18
1	9/17	47
2	8/21	83
2	9/5	10
2	9/11	13
2	9/17	13
3	8/21	90
3	9/5	15
3	9/11	4
3	9/17	11
4	8/21	91
4	9/5	9
4	9/11	9
53	9/8	77
53	9/12	60
55	9/8	113
55	9/12	69
70	9/12	19
70	9/14	12
71	9/12	20
71	9/14	5
Winter 1976		
3	4/5	5.4
3	4/9	4.3
3	4/10	0.7
14a	4/5	12
14a	4/18	<0.2

Table 6 (continued).

Station No. summer 1976	Water samples	
	Date	V_{max}*
72	8/23	85
72	8/24	41

	Sediment Samples	
Summer 1975	Date	V_{max}*
1	9/5	1.51
1	9/17	0.61
2	9/5	0.13
2	9/11	0.13
2	9/17	0.77
3	9/5	0.07
3	9/11	0.02
3	9/17	<0.02
51	9/8	0.78
51	9/13	0.80

*V_{max} values in water samples are reported as ng glutamic per liter per hour and V_{max} values in sediments are reported as μg glutamic acid per gram dry weight per hour.

Table 7. Data summary of measurements made in samples collected in the Barrow and Prudhoe Bay areas.

		Barrow			Prudhoe Bay		
Measurement	Units	n	$\bar{Y}$	s	n	$\bar{Y}$	s
		Water					
V_{max}[*]	ng/l/h	37	37	34	13	44	28
Percent respiration[*]	%	37	62	7	13	54	5
Temperature (*in situ*)	°C	37	1.2	1.5	13	1.0	1.1
Salinity	o/oo	37	22.7	3.1	13	16.7	4.4
pH		37	7.9	0.1	13	7.9	0.1
Bacterial concentration[+]	10^5 cells/ml	37	4.4	2.9	13	4.5	2.7
		Sediment					
V_{max}[*]	µg/g dry wt/h	15	0.40	0.37	18	0.83	0.40
Percent respiration[*]	%	14	41	11	18	32	7
pH		3	7.2	0.1	11	7.5	0.1
Bacterial concentration[+]	10^8 cells/g dry wt	15	4.4	5.3	18	9.2	12.0

Note: n - number of observation; $\bar{Y}$ = mean value; s = standard deviation.

[*] These measurements were made using ^{14}C labeled glutamic acid.

[+] Bacterial concentrations estimated using direct observations with epifluorescent microscopy.

A somewhat different result was obtained from similar measurements made on sediment samples taken in these two regions. The only factors that showed a significant difference in the mean values were the pH and the relative microbial activity (V_{max}). Both the mean values for relative microbial activity and pH were higher in the Prudhoe Bay sediments. The mean bacterial concentration was about twice as high in the Prudhoe Bay area but the variability was high and a statistical analysis of the data revealed that the difference was not significant. The difference seen in the bacterial concentrations did, however, reflect the same difference seen in the V_{max} values when these two regions were compared. In general, the values presented in Table 7 were probably representative of values normally found in the bay waters of the Beaufort Sea in late summer months.

In most cases, the nearshore water samples taken farthest from shore showed the lowest V_{max} values. This is best illustrated in the data collected in the Prudhoe Bay area (Table 8). A comparision was made between the distance of the sampling site from shore and the V_{max} value observed in the water sampled at that site. On the three sampling dates for which there are sufficient data to make a valid comparison, there was a negative correlation between the distance from shore and the V_{max} values observed at that distance. This same trend, however, was not seen in samples taken along the offshore transects sampled in the winter and summer of 1976 (Fig. 17). The sediment samples collected during the September 1977

Table 8. V_{max} measurements in water samples taken at stations in Prudhoe Bay.

Date	Station No.	V_{max}^*	Distance from shore km	Correlation coefficient
9/8	53	77	0.4	-0.39
"	55	113	2.1	
"	55	65^+	2.1	
"	51	38	2.0	
"	54	78	0.8	
9/12	53	60	0.4	-0.85
"	55	69	2.1	
"	56	28	4.0	
"	70	19	5.4	
"	71	20	7.8	
9/13	50	48	0.7	
"	52	32	2.4	
9/14	57	29	4.2	
"	70	12	5.4	
"	71	5	7.8	-0.95
"	72	20	4.3	
"	73	40	2.6	

$^*V_{max}$ values reported as ng glutamic acid taken up per litre per hour.

$^+$Sample taken at a depth of 2 m, all others taken at the surface.

cruise showed higher relative microbial activities in the area near Point Barrow than the areas to the east (Fig. 20). During the August 1978 cruise, we were able to sample a more comprehensive sampling grid (Fig. 19). These data show that the relative microbial activity was highest in the areas near the major rivers along the North Slope. This phenomenon was observed in both water and sediment samples (Figs. 21 and 22). In addition, we observed reduced respiration percentages in the waters adjacent to these rivers (Fig. 23). The patterns of uptake and respiration in the waters were essentially the same as those observed in the Cook Inlet (described in Section I) and in the Norton Sound (described in Section III).

3. Comparison of relative microbial activity in Beaufort Sea with other regions.

Using V_{max} values, we have shown that the relative microbial activity in Beaufort waters was roughly equivalent to that observed during the 1976 Cook Inlet cruise and during the NEGOA cruise the same year (Table 1, Section I). With the exception of the unusually high activity observed in NEGOA sediments, the same can be said for relative microbial activity in the sediments as well.

Comparisons of relative microbial activity can also be made in the studies in which glutamate uptake at one concentration was used (Table 9). If uptake rates in both water and sediments in the Beaufort Sea are compared with data collected in both Cook Inlet (including Shelikof Strait) and Norton Sound, the rates are very similar. With the exception of the two water samples collected in Elson Lagoon in April, 1978, all of the mean values are within the

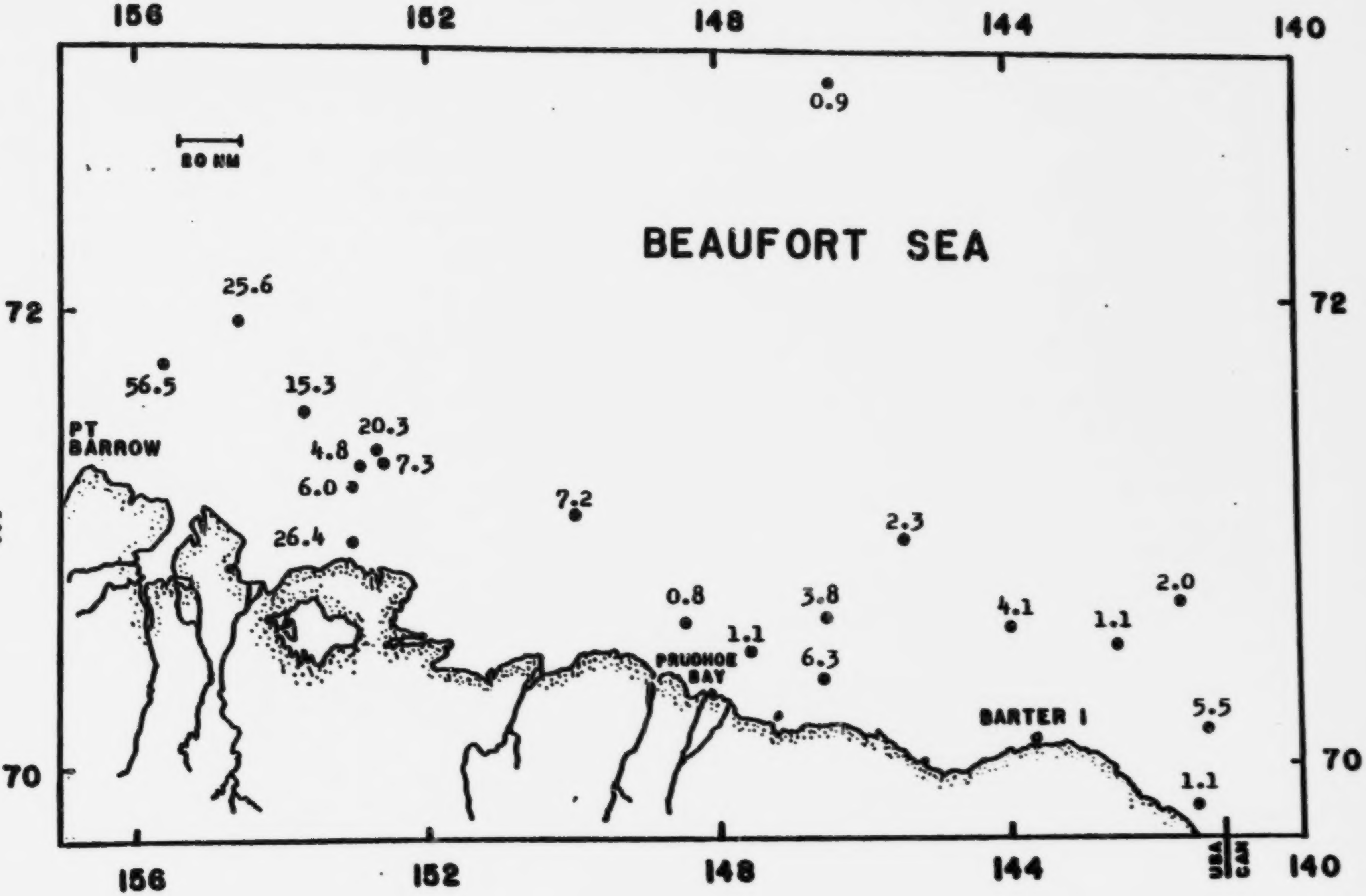
156
152
148
144
140
20 NM
BEAUFORT SEA
72
70
PT BARROW
PRUDHOE BAY
BARTER I
USA
CAN
0.9
25.6
56.5
15.3
20.3
4.8
7.3
6.0
26.4
7.2
2.3
0.8
3.8
1.1
6.3
4.1
1.1
2.0
5.5
1.1

Figure 20. Relative microbial activity in sediments during the September 1977 cruise. The units are ng glucose/g dry wt./h.

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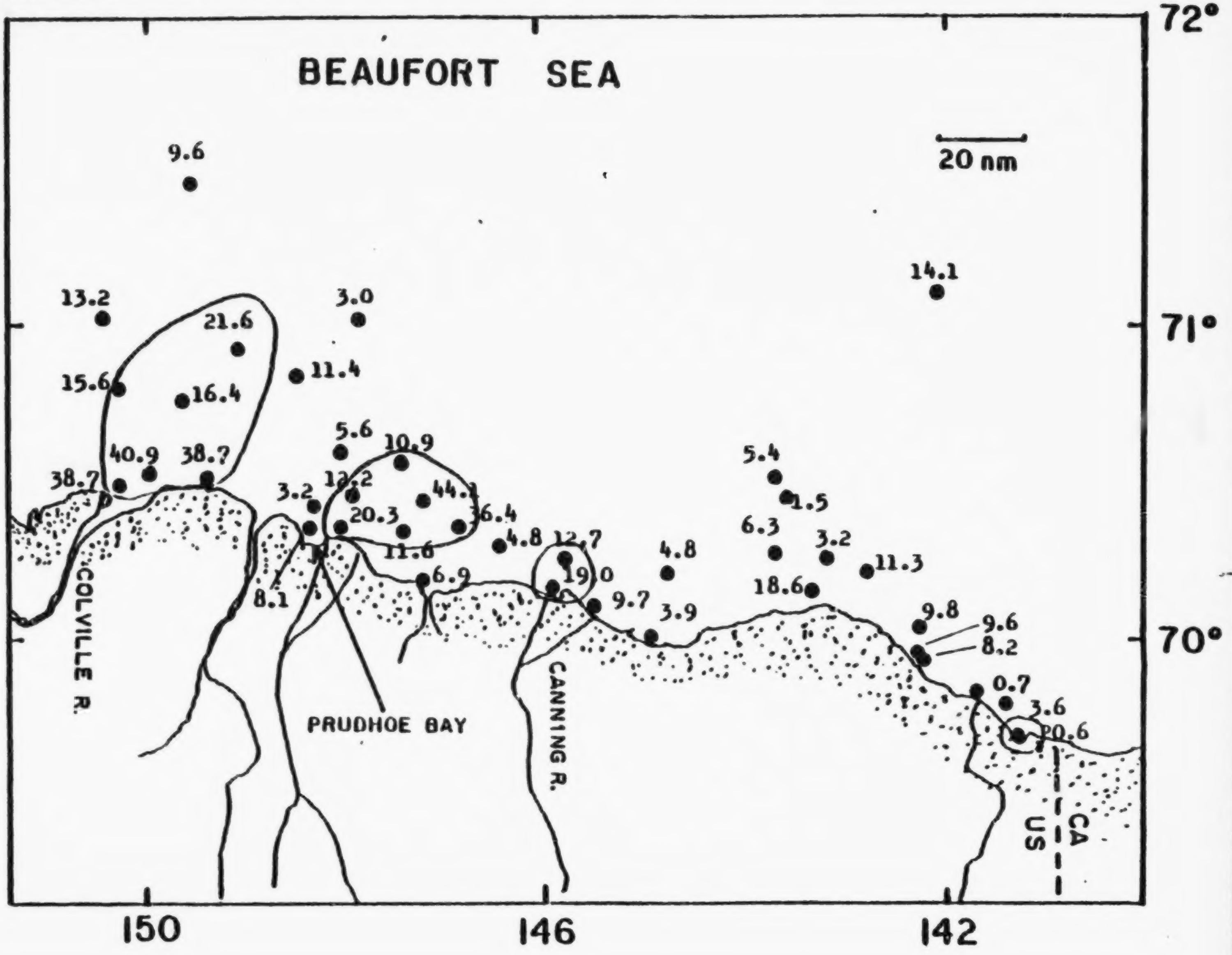

BEAUFORT SEA
20 nm
72°
71°
70°
150
146
142
9.6
13.2
21.6
3.0
11.4
15.6
16.4
14.1
5.6
10.9
40.9
38.7
38.7
3.2
12.2
44.1
5.4
1.5
20.3
36.4
6.3
3.2
11.6
4.8
12.7
4.8
11.3
8.1
6.9
19.0
18.6
9.7
3.9
9.8
9.6
8.2
0.7
3.6
20.6
COLVILLE R.
PRUDHOE BAY
CANNING R.
US
CA

Figure 21. The rate of glutamic acid uptake in water samples collected in August 1978. The units are μg/liter/h.

503

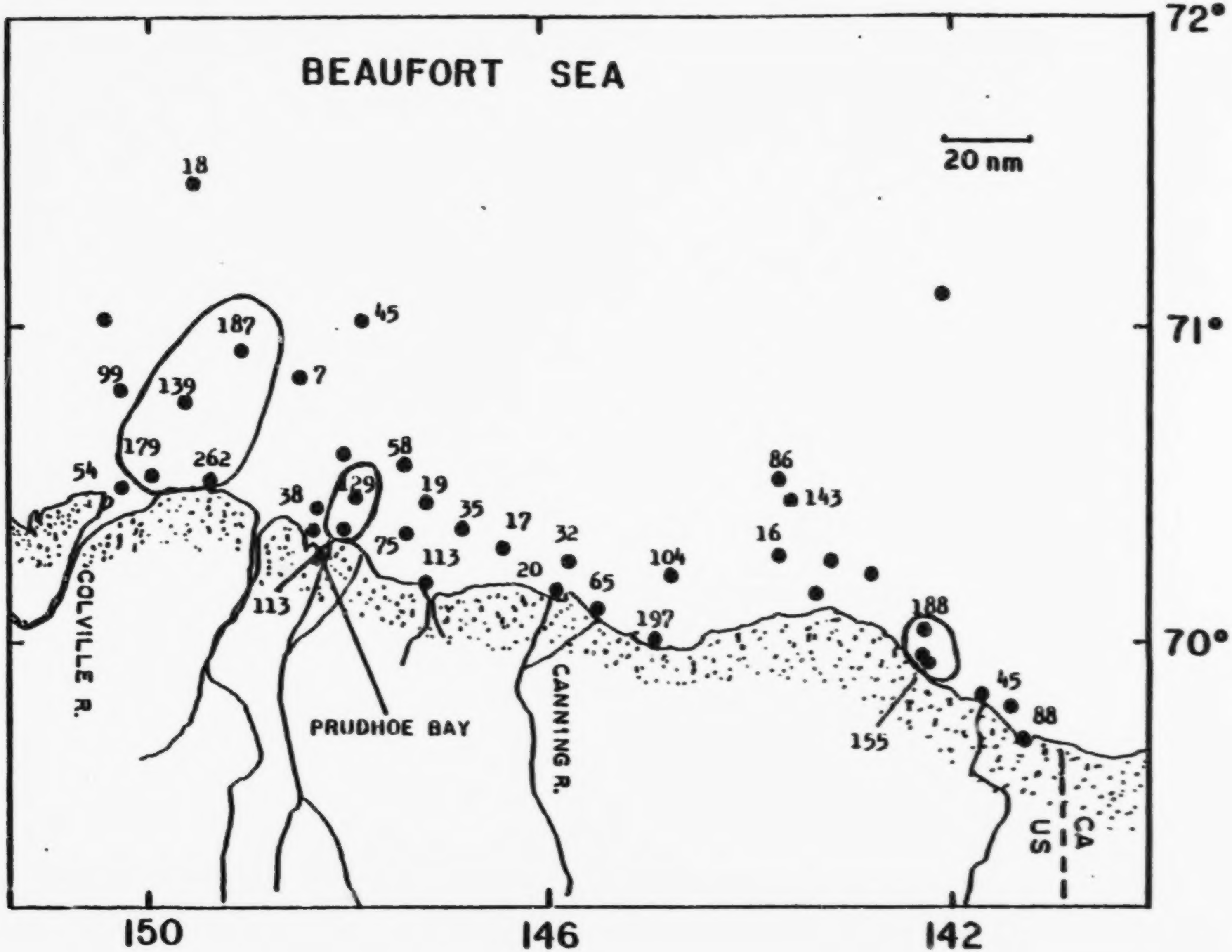
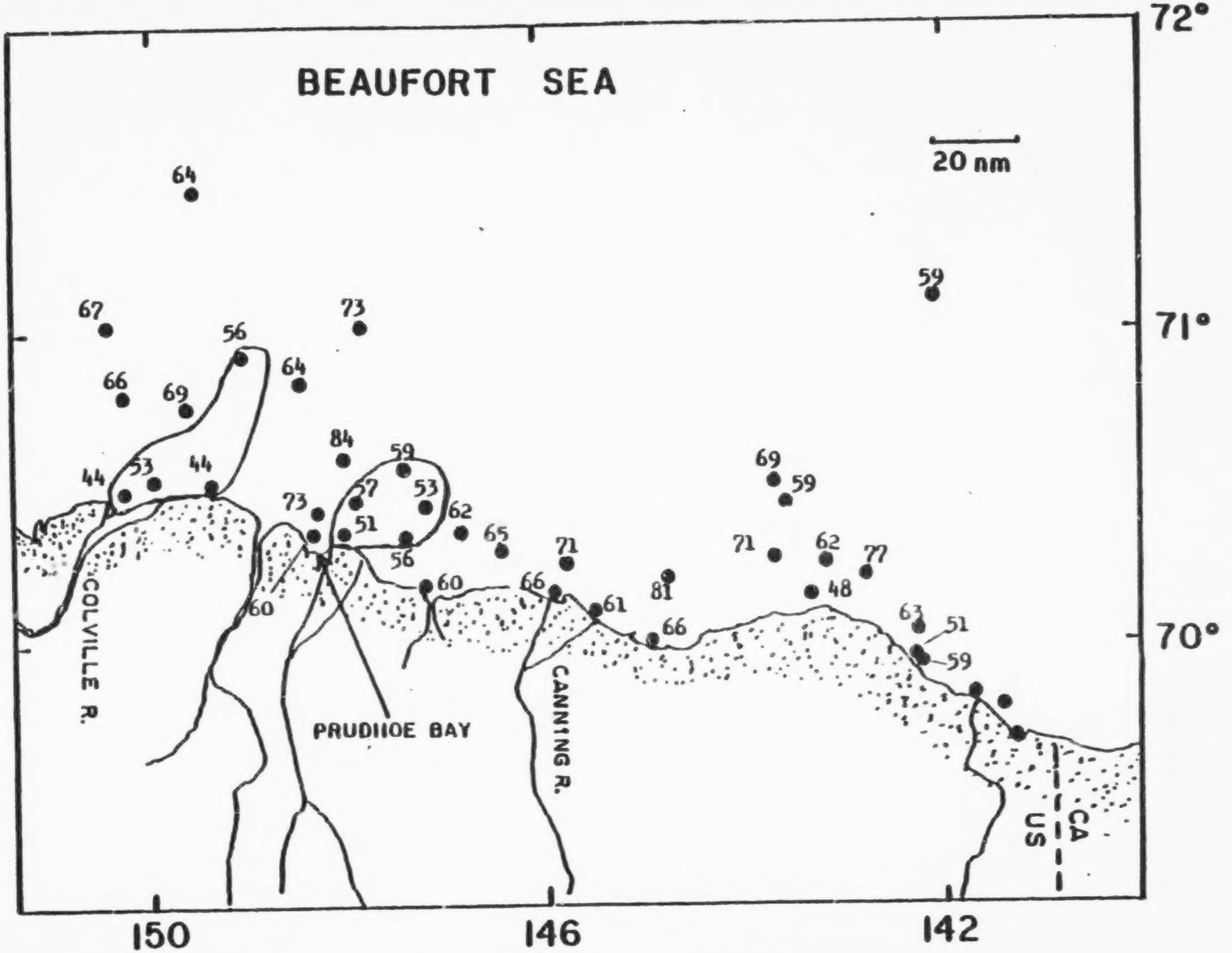

BEAUFORT SEA
20 nm
72°
71°
70°
150
146
142
18
187
45
99
139
7
179
262
54
58
129
38
19
35
17
32
75
113
113
20
65
104
197
86
143
16
188
155
45
88
COLVILLE R.
PRUDHOE BAY
CANNING R.
CA
US

Figure 22. Glutamic acid uptake in sediments collected in August 1978, expressed as ng/g dry wt./h.


BEAUFORT SEA
20 nm
72°
71°
70°
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COLVILLE R.
PRUDHOE BAY
CANNING R.
CA
US

Figure 23. Glutamic acid percent respiration in water samples collected in August 1978.

same order of magnitude. With the exception of the January Beaufort Sea sediment samples, the same can be said for the relative microbial activities in the sediments as well.

Table 9. Comparison of glutamate uptake activity in field station waters and sediment samples.

Area		Water (ng/l/hr)	Sediment (ng/g/hr)
Beaufort Sea	Aug 1976	8	80
	Aug 1978	14	96
Beaufort Sea, Elson Lagoon			
	Jan 1978	4	14
	Apr 1978	0.5	480
	Aug 1978	20	85
	Jan 1979	2	8
Cook Inlet and Shelikof Strait			
	Apr 1977	16	217
	Nov 1977	5	64
	Apr 1978	13	86
Norton Sound	July 1979	19	154

Unfortunately, there is very little data in the current literature with which we can compare the Beaufort Sea data. The data that is available from other geographical regions (Table 10), indicate that the rates observed in the Beaufort Sea during the summer 1975 study was roughly equivalent to that reported by others.

4. The influence of freshwater input on cell concentrations and microbial activity.

One of the many unique characteristics of the Beaufort Sea is the wide range of salinities encountered. During our studies, the widest salinity range observed was seen during the 1976 summer cruise where surface and near-surface samples showed salinities ranging from 5 to 21%. This salinity variation in the area of study was the result of freshwater coming from melting of the pack

Table 10. Comparison of reports of relative microbial activity in marine water and sediment samples taken in various geographical areas. All measurements were made using ^{14}C labeled glutamic acid. Except as noted, the same methods were used for all studies.

Ocean	V_{max}*	No. of samples	Investigation
	Water samples		
Antarctic	10.9	23	Morita _et_ _al_. (1977)
Antarctic	11.2	8	Gillespie _et_ _al_. (1976)
Tasman Bay New Zealand	40.0	1	Gillespie (unpublished)
Eastern Tropical Pacific+	15.2	10	Hamilton and Preslan (1970)

*V_{max} values are reported as ng glutamic acid taken up per unit of sample per hour. The unit of sample in sediments is 1 g dry weight.

+No CO_2 data included.

ice. As a result, the surface-water salinity was much lower than that found at 5 m and below. Seawater samples that were diluted 50% with sterile fresh water showed no significant change in the ability of the natural microflora to utilize glutamic acid even though marked changes in salinity resulted in this treatment. Studies were also made on the relative microbial activity seen in melted ice in contrast to the same measurement made in the water adjacent to the ice (Table 11). Ice when melted showed a high degree of variability which was undoubtedly due to its diverse origin. The microbial activities and bacterial concentrations in the surface water and melted ice were similar. When seawater and melted ice were mixed 1:1 there was no adverse effect on glutamic acid utilization. These observations were further supported by relative microbial activity measurements made at the surface at 15 m (Table 12). The input of fresh water from melted icepack at the surface did not reduce the concentration of bacteria present nor did it adversely affect the uptake of glutamic acid when these values were compared with samples taken from more saline waters at 15 m.

5. Seasonal variations.

The two 1976 Beaufort Sea studies were designed to compare relative microbial activities and bacterial cell concentrations in water and sediment samples taken in the summer and winter at the same stations. Additional data were obtained from other stations during the 1975 summer. The average V_{max} values observed in both water and sediment samples during the winter study were about one

Table 11. Relative microbial activity (V_{max}) and cell concentrations in samples of melted ice, associated seawater, and 50/50% mixtures of the two.

Sample No.	Sample type	V_{max}	10^5 cells/ml	Salinity o/oo
		Summer 1975		
82*	Ice	1	-	1.2
83*	Ice	22	-	0.5
84*	Ice	9	-	2.0
81	Water	24	-	25.0
81 + 84	Mixture	22	-	13.5
†		16		
93*	Ice	34	-	9.0
94*	Ice	68	-	6.0
95	Water	28	-	26.0
93 + 95	Mixture	57	-	17.5
†		31		
		Winter 1976		
BI 120	Ice	4.1	1.3	-
BW 120	Water	2.1	1.2	25.5
BI 122	Ice	2.0	1.6	-
BI 122	Water	2.1	2.7	29.0
BW 123	Ice	0.7	2.2	-
BW 123	Water	4.7	1.3	28.0

*Float ice collected at one location.

†Theoretical result of mixing V_{max} values reported as ng glutamic acid per liter per hour.

order of magnitude lower than that seen in either summer study (Table 13). The data presented in Table 14 illustrate this trend in water samples taken at the same stations during these two seasons. The bacterial concentrations also decreased in the winter but the decrease was not as dramatic as that seen in the V_{max} data. However, there was very little difference seen between the cell concentrations observed in the 1975 summer sediments taken from the inshore stations and the values observed in the 1976 winter sediments taken from the offshore stations even though a 10-fold difference was seen in the V_{max} values in the same samples.

Table 12. The V_{max} values and cell concentrations in surface waters and water samples taken at 15 m at four stations in the Beaufort Sea.

	Surface water		Water at 15 m	
Station No.	V_{max}*	10^5 cells/ml	V_{max}*	10^5 cells/ml
24	6	3.2	6	2.3
23	27	4.6	5	4.5
22	6	3.0	0.4	1.9
15a	7	3.7	2	5.4

*V_{max} values are reported as ng glutamic acid per liter per hour.

Table 13. Data summary for observations made on all samples taken in the Beaufort Sea.

		Summer 1975			Winter 1976			Summer 1976		
Measurement	Units	n	$\bar{Y}$	s	n	$\bar{Y}$	s	n	$\bar{Y}$	s
V_{max}* (water)	ng/l/h	50	44	30	21	3.1	3.7	16	21	24
V_{max}* (sediments)	mg/g dry wt/h	33	0.61	0.47	14	0.06	0.04	11	0.83	1.20
Percent respiration (water)*	%	50	59	7	23	85	14	16	46	11
Percent respiration (sediment)*	%	33	37	4	14	39	13	11	23	6
Bacterial concentration (water)+	10^5 cells/ml	50	4.5	2.7	23	1.5	0.5	18	3.7	1.3
Bacterial concentration (sediment)+	10^8 cells/g dry wt	33	6.6	8.9	13	10	6.7	11	106	7.9
Salinity	o/oo	50	20.5	5.0	23	24	24	18	15.3	4.7
Temperature	°C	50	1.2	1.0	23	-1.9	-1.9	18	-0.1	0.3

Note: n = number of observations; $\bar{Y}$ = mean value; s = standard deviation.

*These measurements were made using ^{14}C labeled glutamic acid.

+Bacterial concentrations estimated using direct observations with epifluorescent microscopy.

Table 14. A seasonal comparison of V_{max} values observed at several stations.

Station No.	V_{max}*		
	Summer 1975	Winter 1976	Summer 1976
2	30†	1.1	-
3	30†	3.5‡	-
15a	-	4.2	7
16	-	2.1	56
21	-	<0.2	4
22	-	0.2	6
23	-	0.2	27
24	-	0.4	6
74	-	2.8	85
80	-	0.7	34

*V_{max} values are expressed a ng glutamic acid taken up per liter per hour.

†Average of four measurements.

‡Average of three measurements.

A statistical analysis of these data shows that there was a significant difference in the V_{max}, percentage of respiration, cell concentration, and salinity between the water samples taken in the winter and those taken during both summers. In the sediment samples, there was a significant difference seen between the V_{max} values during the same sampling seasons. The cell concentrations in the sediments were significantly lower in the winter as compared to the summer 1976 measurements but they were not significantly different from those observed in the summer of 1975.

The differences seen in the microbial activity in the sediment and surface-water samples during the summer and winter studies probably reflect variations in nutrient availability during these two seasons rather than changes in water temperature. The widest difference in the average surface-water temperature observed between winter and summer conditions was about 3°C (Table 13). A study was conducted to determine the effects of incubation temperature on four water samples taken in the Barrow area during the 1975 summer study (Fig. 24). These data indicate that a drop in temperature of 3°C will decrease the V_{max} value by about 50%.

In addition to these data, we also collected seasonal data during the Elson Lagoon study (Table 15). In these studies, we compared the relative microbial activity in water and sediment samples using single concentrations of glucose and glutamic acid. The microbial activity in the water samples was low during the January and April sampling periods but much higher during August. In the sediments, the activity was lowest in January and highest in April and August.

6. Comparison of microbial activity in sediments and in the water column.

The sediments of shallow waters have long been recognized as an important area of microbial activity in marine ecosystems. It has also been known that microbial activity in sediment is much higher than that found in water. This trend was also observed in this study (Table 13). In the summer 1975 study, the average value for the potential rate of glutamic acid uptake and mineralization was 0.52 μg glutamic acid per gram dry weight of sediment per hour. There is no way to compare directly this figure to potential activity in a comparable volume of water but a reasonable approximation can

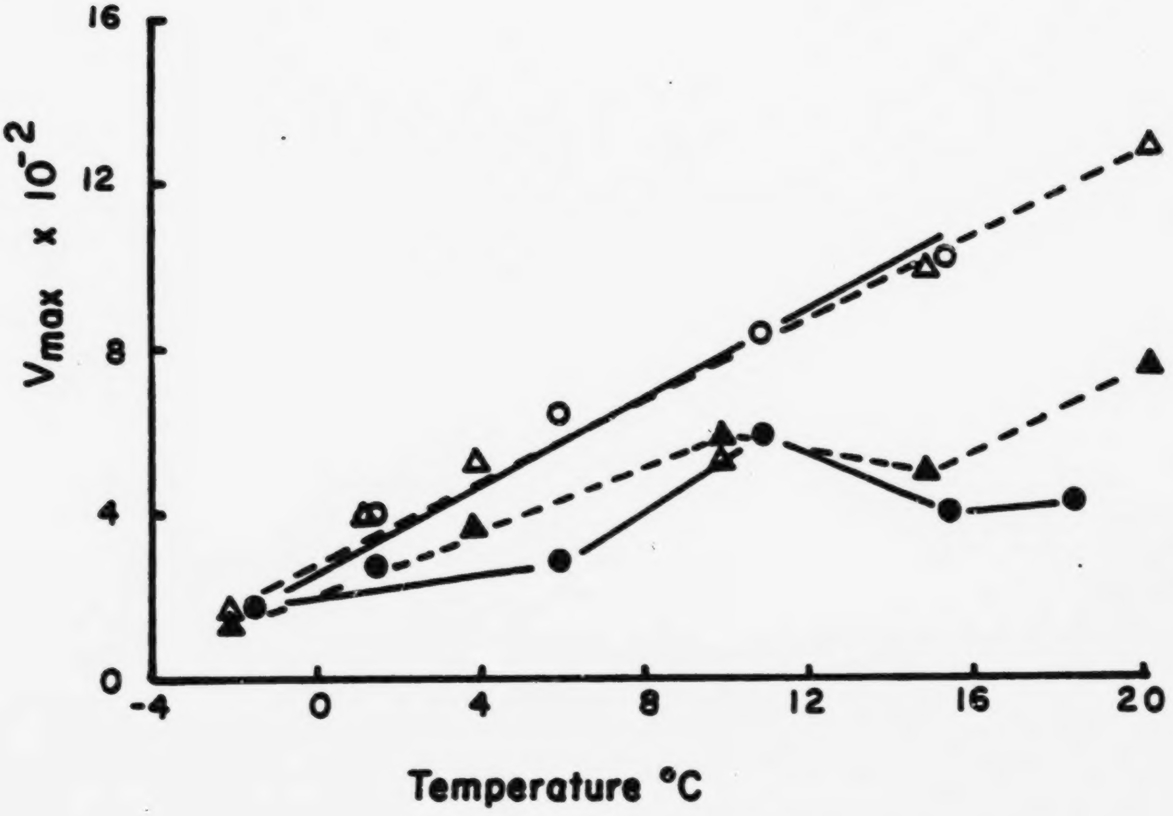

Figure 24. The effect of incubation temperature on the maximum velocity of glutamic acid uptake (V_{max}) in natural microbial populations of water samples taken at stations 5a and 5b. (△)(○), measurements made on samples taken at station 5a. (▲)(●), measurements made on samples taken at station 5b. V_{max} values reported as ng glutamic acid taken up per litre per hour, in units of 10.

Table 15. Seasonal comparison of microbial uptake and nitrogen fixation rates in water and sediment samples from Elson Lagoon.

	WATER		SEDIMENTS		
Jan 1978	Glucose[1]	Glutamate[1]	Glucose[2]	Glutamate[2]	N_2fix[3]
401	0.4	4.8	-	-	-
401 ice	0.5	4.5	-	-	-
402	0.3	3.8	3.4	22.6	0.15
403	0.3	3.4	7.8	27.6	0.15
407	-	-	0.2	2.8	0.10
408	0.3	2.4	1.2	6.0	0.11
Mean	0.4	3.8	3.1	15	0.13
Apr 1978					
501	0.3	0.6	-	-	
502	0.3	0.4	-	-	
503	0.1	-	12.2	479	
Mean	0.2	0.5	[6.2][4]	[234][4]	[1.4][4]
Aug 1978					
601	15.6	34.2	2.7	19	1.1
604	2.3	4.6	-	-	-
606	433.9	21.6	-	-	-
607	-	-	13.2	161	0.3
Mean	21	20	8	90	0.7
Jan 1979					
701	0.2	0.6	-	-	-
702	0.2	2.7	-	-	-
703	-	-	2.4	7.0	-
704	-	-	3.7	7.9	
Mean	0.2	1.7	3.1	7.5	-

1 - ng/l/hr

2 - ng/g dry wt/hr

3 - ngN fixed/g dry wt/hr

4 - mean of 5 control and oiled trays

be made. A relative comparison between microbial activity in sediments and seawater can be made by contrasting the activity in a given volume of seawater with an equal volume of sediment-seawater slurry. In the summer 1975 study, the average V_{max} in the undiluted sediment slurries was 2.5×10^2 μg per liter per hour. This is roughly four orders of magnitude higher than the average figure of 3.8×10^{-2} μg observed in the water samples.

Another comparison of potential microbial activity in sediments and seawater can be made by comparing the total activity in an average water column with activity in the underlaying sediment. To make such a comparison, at least two assumptions must be made. The first is that most of the microbial activity is taking place in the first 2 cm of the sediment. This assumption is based on the findings of ZoBell (1942) and others who have shown that the vast majority of bacteria in sediments are found in the top 2 cm. The other assumption is that diluting the sediment sample with sterile artificial seawater does not significantly affect the resulting observed activity.

Keeping these assumptions in mind, the potential rate of glutamic acid utilization in the average water column (1 m^2 x 3 m deep) can be compared with the same potential in the sediment below that square meter of water. The maximum potential for glutamic acid utilization in an average water column in the test area was 0.1 mg glutamic acid in the water column per hour. If our assumptions are correct, the sediments in the water columns studied in the inshore stations (summer 1975) had, on the average, 400 times greater activity than the entire overlaying water column. This figure should be considered an underestimation of what is probably the true value because: (1) we have assumed that there is no microbial activity in the sediments below 2 cm, (2) the value was calculated in terms of a slurry which contained roughly 50% seawater, (3) the actual sediment samples studied contained material to a depth of 4 cm.

7. Adjustment of microbial populations to hydrocarbons in the environment.

During the course of our studies, we have collected two sets of data which shows that benthic microbial populations may be changing in response to the inputs of hydrocarbons into the Beaufort Sea. An analysis of biodegradation potentials in sediments collected during the September 1977 cruise indicates that the benthic microbial populations in sediments to the east of Prudhoe Bay have the highest potential for degrading crude oil (Fig. 25). Sediments collected during the August 1978 cruise shows that the benthic microorganisms in this same general area are affected the least by the addition of crude oil on a short-term basis (Fig. 26). In these studies, the lack of a reduction in the amount of glucose taken up by the microorganisms in the presence of crude oil can be interpreted as an indication of prior exposure.

B. Rates of nitrogen fixation in Beaufort Sea sediments.

1. Geographical distrubution.

We measured rates of nitrogen fixation in two sets of sediments during two cruises in the Beaufort Sea (Figs. 27 and 28). The set

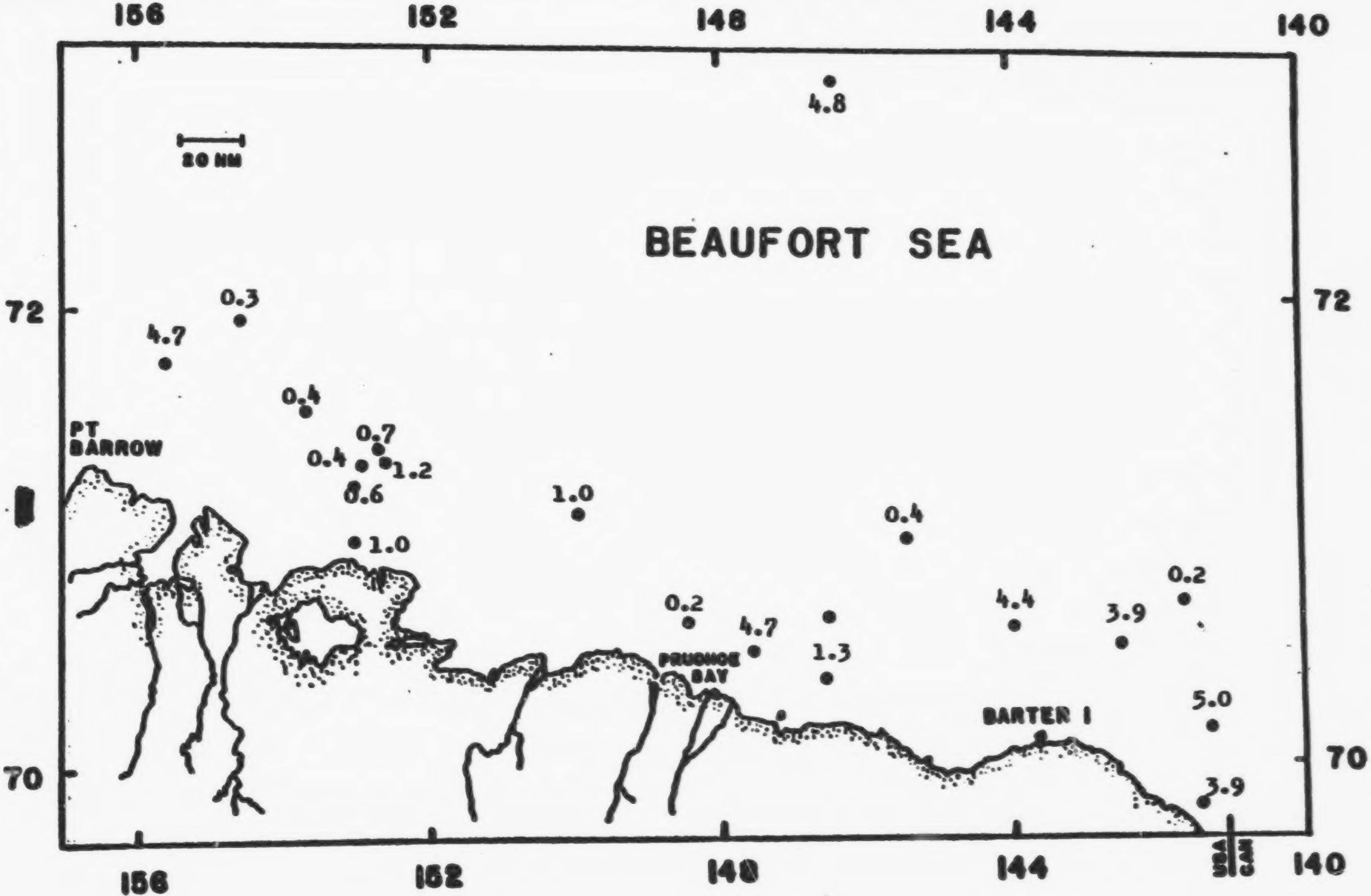
156
152
148
144
140
72
70
20 NM
BEAUFORT SEA
PT BARROW
PRUDHOE BAY
BARTER I
4.8
0.3
4.7
0.4
0.7
0.4
1.2
0.6
1.0
1.0
0.4
0.2
4.7
1.3
4.4
3.9
0.2
5.0
3.9

Figure 25. Crude Oil biodegration potentials in sediments collected during September 1977, expressed as thousand DPM/g dry wt.

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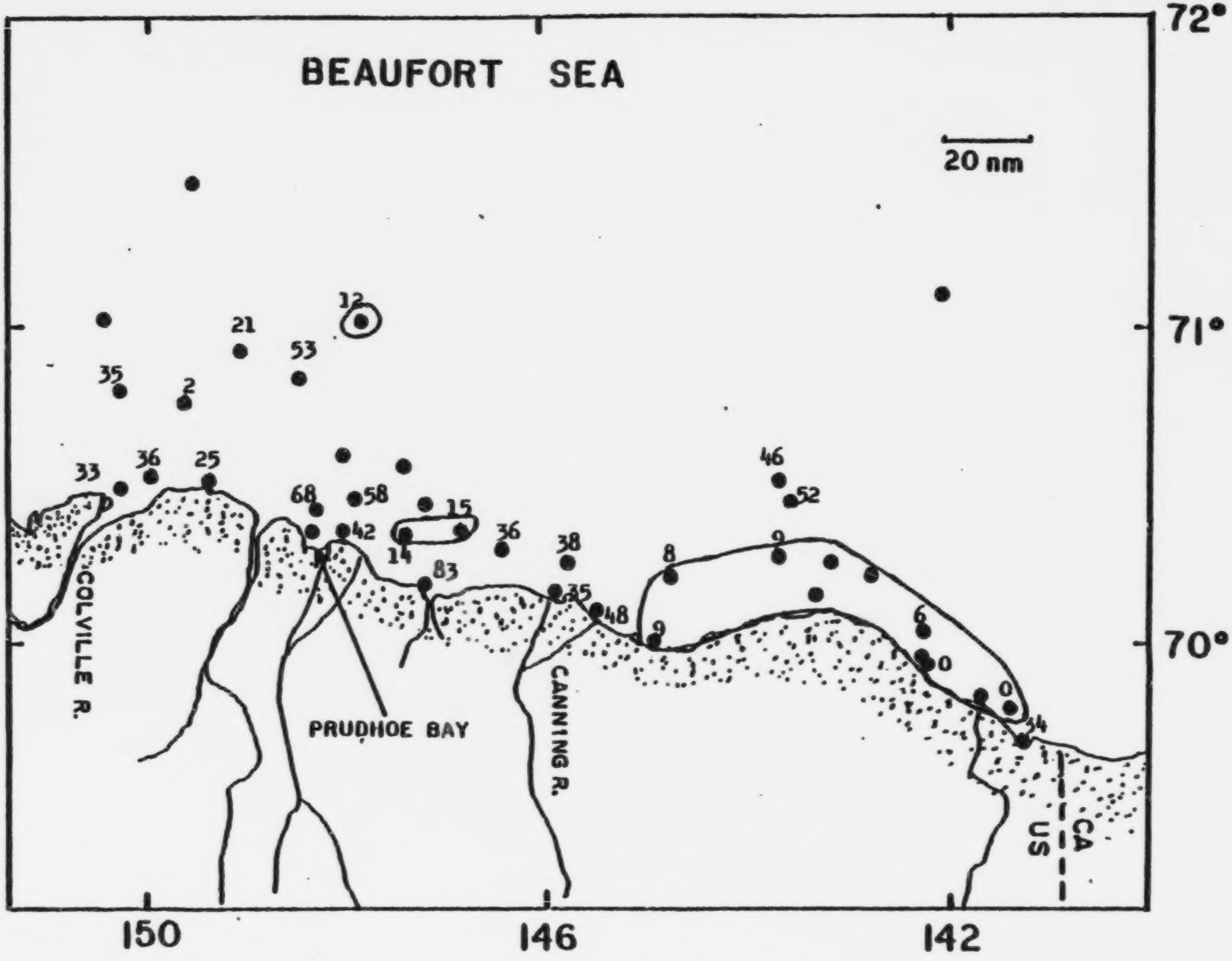
BEAUFORT SEA
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52
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34
COLVILLE R.
PRUDHOE BAY
CANNING R.
US
CA

Figure 26. Percent reduction in the rate of glucose uptake in sediment samples exposed to Prudhoe crude oil when compared to nonoiled samples. Sediments were collected during the August 1978 cruise.

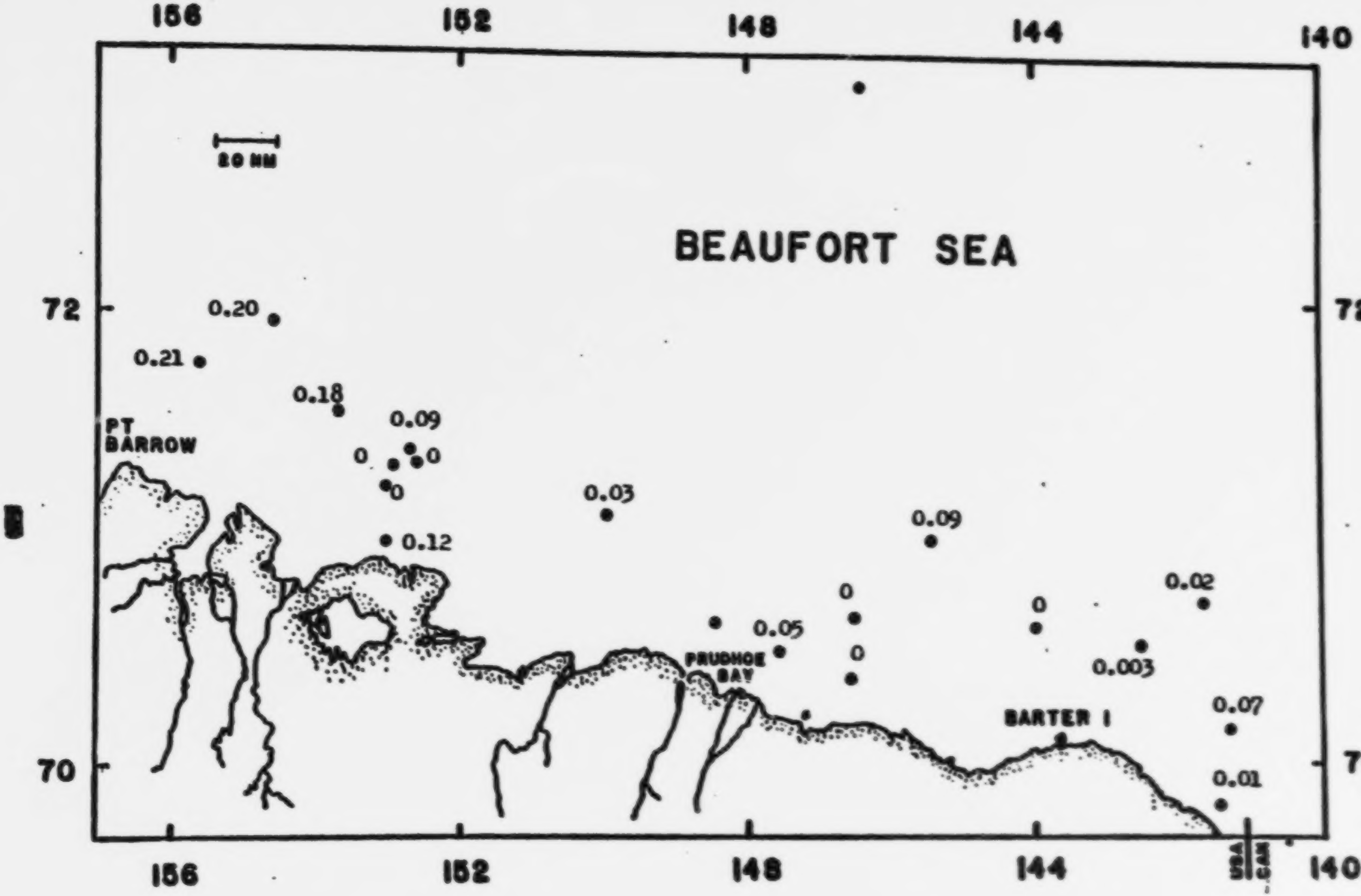
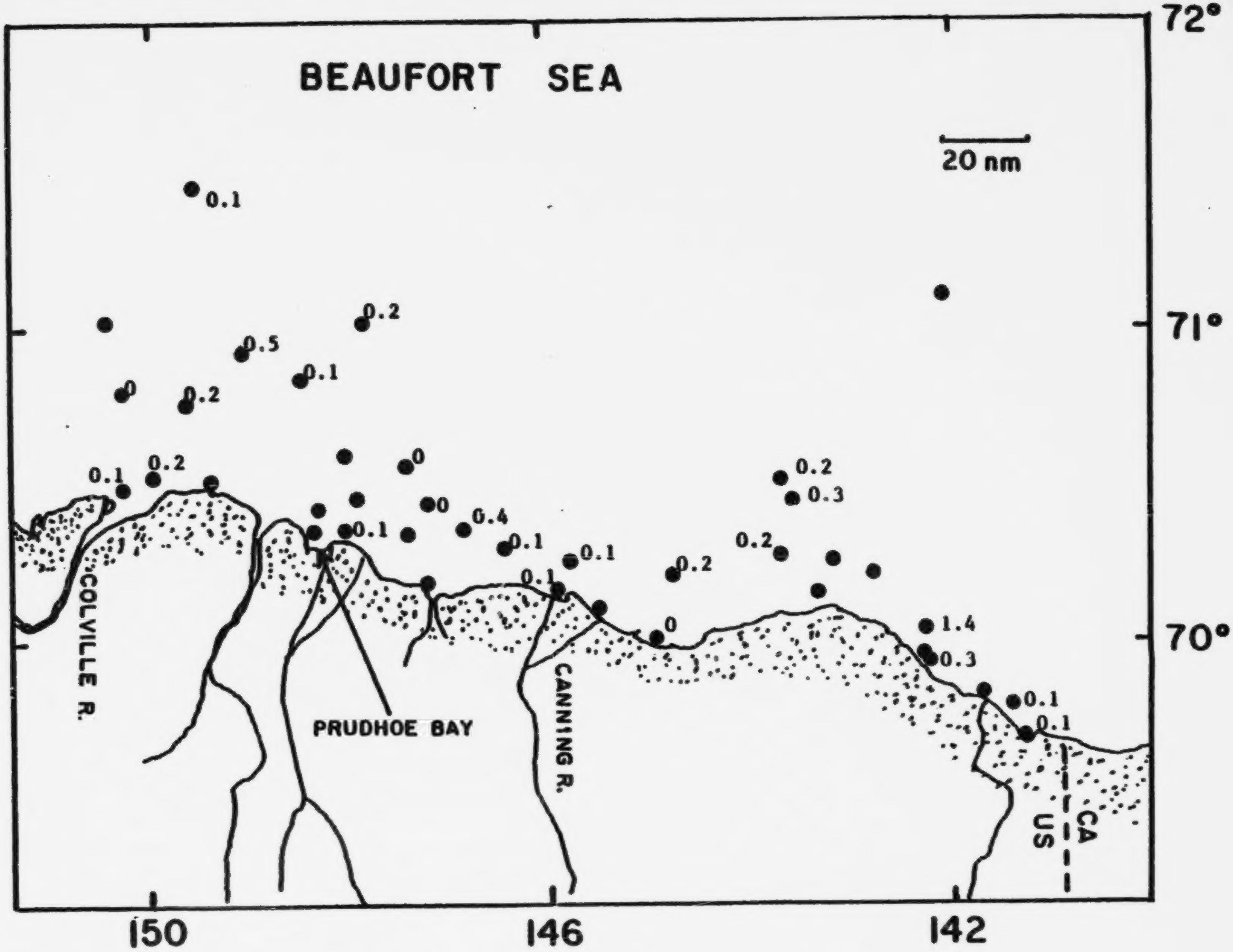

156
152
148
144
140
20 NM
BEAUFORT SEA
72
70
0.20
0.21
0.18
0.09
PT BARROW
0
0
0
0.03
0.12
0.09
0.02
0
0
0.05
0
0
0.003
PRUDHOE BAY
0.07
BARTER I
0.01
USA
CAN

Figure 27. Nitrogen fixation rates in sediments collected during the September, 1977 cruise, expressed as ng N_2 fixed/g dry wt./h.


BEAUFORT SEA
20 nm
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0.2
0.5
0.1
0
0.2
0.1
0.2
0
0
0.4
0.1
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0.1
0.1
0.2
0
0.2
0.3
0.2
1.4
0.3
0.1
0.1
COLVILLE R.
PRUDHOE BAY
CANNING R.
US
CA
72°
71°
70°
150
146
142

Figure 28. Nitrogen fixation rates in sediments collected during the August 1978 cruise, expressed as ng N_2 fixed/g dry wt./h.

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of data that was collected in September 1977 covered the largest geographical area. In this study, the highest rates were observed in sediment samples collected near Point Barrow. The nitrogen fixation rates to the east were generally lower. During the second cruise which included the area between the Colville River and the Canadian-US border, there were no significant geographical trends noted. If one compares these data with similar data collected in Cook Inlet, Shelikof Strait and Norton Sound, there were differences noted in the mean values (Table 4, Section I). The mean nitrogen fixation rate observed in Norton Sound was not significantly different from that observed in the Beaufort. Generally the rates observed in Cook Inlet were higher but the differences were not statistically significant. The mean values for nitrogen fixation in Shelikof Strait were significantly higher than those observed in the Beaufort Sea.

2. Seasonal differences.

There has been only one study in which we have collected nitrogen fixation data on a seasonal basis in the Beaufort Sea. We measured nitrogen fixation rates in sediments located near the oiled tray experiment in Elson Lagoon (Table 15). Three sets of observations were made; January 1978, April 1978 and August 1978. The lowest rates were observed in January 1978; intermediate values in August and the highest value in April. The August figure was approximately 1/3 of the mean observed in sediments analyzed during the NorthWind cruise later that same summer. Unfortunately, there was one non-oiled sediment analyzed in April 1978. In all, there were five sediment samples analyzed (one control and four oiled). The mean value for all five was 1.3 ng x g^{-1} x h^{-1}. This value is exactly one order of magnitude higher than the rate observed in January 1978.

C. Total adenylates in Elson Lagoon sediments.

The combined concentrations of ATP, ADP, and AMP was measured in the sediments of Elson Lagoon in August 1978 and January 1979. The total concentration of these three chemical species (total adenylates) was higher in the summer than in the winter. From this information, we were also able to calculate energy charge. The higher the energy charge, the more metabolically active the population is. The mean energy charge in summer sediments was 0.97 and in the winter it was 0.25 (Table 41, Section IV).

D. Crude oil toxicity studies.

During the August 1978 Beaufort Sea cruise, we measured the acute toxic effects of Prudhoe Bay crude oil on nitrogen fixation rates and relative microbial activity in both water and sediment samples. During the Elson Lagoon oiled tray experiment, we measured these variables and the long-term effects of crude oil on these variables. The results of these studies are reported and analyzed in Section IV of this report.

E. Data storage.

A complete data set from the Beaufort Sea is located in the following NIH data files:

	#
Summer 1975	188
August 1976 Glacier cruise	236
September 1977 " "	307
September 1978 Northwind	309

V. Discussion

A. Geographical differences.

When one compares the relative microbial activity observed in the Beaufort Sea with similar observations made by us and others in different regions, the rates are very close to rates measured elsewhere. This is true even in regions where the mean water temperature is much higher. This suggests to us that the microbial populations in the Beaufort Sea are very well adapted to functioning in this extreme environment. This supposition is supported by data collected by Dr. Atlas (RU #29). He consistently finds more bacterial strains growing on agar plates incubated at 4 C than those incubated at 15 C when he innoculates his plate with seawater or sediments collected in the Beaufort Sea. Thus it cannot be assumed that metabolic processes are inherently slow in the Beaufort Sea because the temperatures are lower than those found in other regions.

Within the Beaufort Sea we did see regional differences in microbial function. The offshore area near Barrow showed higher levels of both nitrogen fixation and microbial activity than the areas to the east. The same pattern was seen when we compared relative microbial activities in the waters and sediments of inshore samples collected in Barrow and Prudhoe Bay during the summer of 1975. The one exception to this is the area near Barter Island. It is quite likely that these variations are related to the sedimentation rates in these areas.

During the Prudhoe Bay study which was conducted landward of the barrier Islands, we found that the relative microbial activity was highest nearshore. In other studies where samples were collected seaward of the barrier Islands along tracks normal to the coast, there was no consistent pattern of microbial activity along those lines. These data suggest to us that nutrients become trapped behind the barrier Islands. Most of these nutrients entering the system are probably from the major rivers as indicated by the data collected during the August, 1978 cruise (Figs. 21, 22, and 23). The patterns of relative microbial activity and respiration percentages observed near the rivers is very similar to those observed in the Cook Inlet and Norton Sound. The region in which this phenomenon is best documented is in the Cook Inlet (see Section 1). In all three areas where there is large plume of freshwater coming into the environment, there is an region where the relative microbial activity is very high and the respiration percentages are very low. We feel that this indicates a significant nutrient input from terrestrial sources. In these regions, the quality and quantity of the nutrients available are such that the microorganisms are growing very rapidly. This is reflected in high metabolic activity and high production of bacterial biomass. This biomass, in turn, represents a major nutrient source for organisms at higher trophic levels.

In total, these geographical trends suggest to us that the areas landward of the barrier islands, particularly near the major rivers, would be the place where crude oil would most profoundly affect microbial function. As we will explain in detail in Section IV, a crude oil spill in these areas could greatly effect the overall productivity since we have shown that long-term exposure of sediments to crude oil can alter overall productivity in several ways.

The other regional patterns that we have observed concern the adaptation of natural microbial populations to the presence of hydrocarbons. We have used two methods to determine if a population of microorganisms had been exposed for an extended period of time to hydrocarbons. The association between prior exposure and the variables that we measure is based on the assumption that a microbial population adjusts to the presence of crude oil. A more detailed explanation is given in Section IV of this report.

If a natural microbial population is exposed to hydrocarbons, then a greater portion of that population is capable of degrading hydrocarbons. The more hydrocarbon degrading bacteria present, the higher the crude oil biodegradation potential. This type of association has been established in the Cook Inlet studies reported by Roubal and Atlas (1978). Our study of Beaufort Sea sediments showed that the biodegradation potentials were highest in the sediments collected to the east of Prudhoe Bay (Fig. 25).

The other method that we have used is the study of the acute effect of crude oil on glucose uptake in Beaufort Sea sediments. Using this approach, we have consistently observed that the microbial populations in both waters and sediments that have not had a prior exposure to hydrocarbons, show reduced rates of glucose uptake in the presence of crude oil (Griffiths et al., 1980b). In samples where there is little or no acute crude oil effect on glucose uptake, it can be assumed that this population has had prior exposure to hydrocarbons. In Cook Inlet, we have observed essentially the same patterns using this technique as those reported by Roubal and Atlas (1978). In the Beaufort Sea, the area where we measured little or no acute effects of crude oil on glucose uptake was to the east of Prudhoe Bay. Since essentially the same results were obtained by two different techniques, we must assume that the benthic microbial populations in this region have been exposed to hydrocarbons. The most likely source of hydrocarbons in this area is crude oil.

B. Seasonal differences.

Whenever we have compared relative microbial activity in Beaufort Sea waters seasonally, we have observed significant differences summer and winter (January and April) rates. In the winter, the activities are typically ten times lower than in the summer. We have observed about the same difference in our seasonal studies in Kasitsna Bay (Section IV). It is also of the same magnitude as the seasonal changes reported by Carney and Colwell (1976) who attributed this seasonal fluctuation to temperature differences. Our study on the effects of incubation temperature on apparent microbial activity (Fig. 24) shows that the seasonal temperature changes could not account for the seasonal differences that we have observed in the Beaufort Sea. If temperature was the only factor involved, we would anticipate a change by a factor of 2 instead of a factor of 10. An alternative explaination, is that nutrients are limiting in the winter. Under winter conditions, there would be very little if any terrestrial input and very little from phytoplankton until the first under-ice phytoplankton bloom which often occurs in April (Dr. Horner, personal communication).

If the observed seasonal differences were due to temperature changes, we would expect to find essentially the same magnitude of change in the microbial activity in sediments. This was not observed. Typically, there was a 2 to 3 fold seasonal change in benthic microbial activity. There is, of course, a larger reservoir of nutrients in the sediments than is the overlaying water column. There are undoubtedly enough nutrients in the sediments during the winter months to keep the microorganisms active during this time. The mineralization of nutrients during the winter months would result in microbial biomass which could be used by benthic organisms as a food source at a time when other food sources are minimal. Busdosh and Atlas (1977) have observed that amphipods are very active during the winter months in Elson Lagoon. It is possible that these and other organisms obtain food (either directly or indirectly) from microbial biomass during these months. Another byproduct of nutrient mineralization by microorganisms is inorganic nitrogen and phosphorous. These inorganic nutrients, which are required by phytoplankton for growth, may accumulate during the winter months as a result of detrital mineralization by bacteria. The build-up of these nutrients in the winter may supply much of the nitrogen and phosphorous required for the spring phytoplankton blooms. We have observed that crude oil adversely effects this mineralization process. It is therefore very likely that if crude oil became incorporated into marine sediments, that the availability of inorganic nutrients for phytoplankton growth may be significantly reduced (see Section IV for details).

A seasonal variation in the percentage of glutamic acid respired was noted in seawater samples. In the winter, the average percentage respiration in the water was higher than that found in the summer. This shift in the percentage respiration could reflect the quality and the quantity of nutrients available to the organisms during these two seasons. During the winter months, the nutrient concentrations are presumably low and there may be deficiencies in growth factors required for biosynthesis. As a result, a larger percentage of the utilized glutamic acid is used for the energy requirements of the cells under nutrient-limited conditions and thus is respired to CO_2 rather than incorporated into cell material.

This concept is supported by the percentage respiration data collected in sediments. As shown in Table 13, the average percent respiration was lower in the sediments than in the water samples during each of the three field-study periods. Nutrients are known to be concentrated in the sediments. As a result, one would expect to find a greater percentage of glutamic acid being incorporated into cell material in bacteria growing in sediments.

Seasonal variations were also observed in nitrogen fixation rates (Elson Lagoon oiled tray experiment). The lowest rates were observed in the month of January. The highest rates observed were in April and intermediate rates were observed in the summer. This is a different seasonal pattern than that observed during the Kasitsna Bay study (Section IV). In this study, the highest rates were observed in November and the lowest rates in July. These differences suggest that there may be basic qualitative and quantitative differences in the types and relative utilization of sediments of these two regions. In other systems that we have studied, an increase in nitrogen fixation rates usually indicates the availability of a readily utilizable nutrient source with a high carbon:nitrogen ratio. It is possible that, in the Beaufort Sea sediments, the conditions that are favorable for nitrogen fixation do not occur until April, whereas in the sediments of Cook Inlet, they take place in the late fall.

One curious aspect of the seasonal data collected during the Elson Lagoon experiment was the unusually high relative microbial activity associated with the control sediment during the April, 1978 field study. Unfortunately, we analyzed only one control during that field trip. If the mean uptake rates are calculated for all five samples (including the oiled samples which usually have rates lower than the control) the values are 6.2 and 234 ng x g^{-1} x h^{-1} for glucose and glutamic acid respectively. When compared to the other seasonal glucose values observed in Elson Lagoon, these are approximately the same as those observed in August, 1978 and it is 2 times greater than that observed in January, 1978 and 1979 (Table 15). The mean value for glutamic acid uptake was greater in April 1978 than at any other time. This increased benthic microbial activity comes at the same time as the highest nitrogen fixation rates. We know from the water data that there are no organic nutrients coming into the sediments via the water column at that time. These data suggest that the detrital food chain is being activated in the spring. This is very different from what we have observed in Kasitsna Bay. The net effect of this activity would be to provide a food source for the benthic community prior to or in conjunction with the spring under-ice phytoplankton bloom. It would also provide some of the inorganic nutrients required for that phytoplankton bloom by mineralization of organic nitrogen and phosphorous.

Another set of variables that show distinct seasonal trends is the total concentration of adenylates and the energy charge that is calculated from the ratios of specific adenylate species (Table 41, Section IV). Since all living things contain adenylates this assay is not specific for microorganisms. The total concentration of adenylates (ATP + ADP + AMP) can be used as an index of total biomass in the sample analyzed. In our experiments, no organisms larger than 2 mm were included. In August 1978, the mean adenylate concentration in the controls was 164 nM x g dry wt^{-1}. In January, 1979 the mean was 15 nM x g dry wt^{-1} which was approximately 10 times less than that observed in August. This is a much greater seasonal variation than that observed in Kasitsna Bay (Section IV).

From the adenylate data one can also calculate an energy charge value which should reflect the metabolic state of the population (Wiebe and Bancroft, 1975). In general terms, the higher the ratio, the more metabolically active the population as reflected by proportionally higher concentrations of ATP. The summer ratio was approximately 4 times greater than that observed in the winter. The seasonal difference in these ratios was also much greater than that observed in Kasitsna Bay. These data lead us to conclude that the overall metabolism of the benthic community slows down much more during the winter in the Beaufort Sea than it does in Cook Inlet.

C. The role of microorganisms in sea ice.

The sea ice plays an important role in the function of the ecosystem in the Beaufort Sea but almost nothing is known about the effects of freezing and thawing seawater on the natural microflora. It was felt that these processes may have a profound effect on nutrient recycling in these waters. The relative microbial activity in the melted ice was compared with microbial activity in the surrounding seawater. It was found that 6 of the 8 ice samples tested showed activities as high or higher than that observed in the associated seawater (Table 11). There are several possible explanations for this observation: (1) the organisms that do survive freezing are the ones that are most actively utilizing glutamic acid, (2) there is a significant number of marine bacteria that survive freezing and are able to grow so that there is not net loss in activity, (3) many bacteria are not actually frozen but are concentrated into highly saline pockets of water within the ice. Of these possibilities, the latter seems the most likely.

It is currently thought that when seawater is frozen, small pockets of highly saline seawater remain throughout the ice. It is quite likely that nutrients and bacteria are also concentrated in these saline pockets. One study has been made of the number of colony-forming units (CFU) found in ice cores taken in Beaufort Sea ice (R. M. Atlas, personal communication). It was found that the bacterial populations showed a high degree of patchiness throughout the core. These data tend to support the above concept. If the bacteria are concentrated in highly saline pockets, the high salinity itself may afford some degree of freeze-injury protection.

Salinity profiles of waters in the Beaufort Sea (taken when there was a significant icepack melting) showed a shallow lens of brackish water at the surface of the water column. These waters showed relative microbial activities and bacterial cell concentrations that were as high or higher than that found in the much more saline waters at 15 m (Table 12). This along with the seawater dilution studies shown in Table 11 suggest that the freshwater input from melting sea ice does not significantly alter the parameters studied.

D. Effects of crude oil and the dispersant Corexit 9527 on microbial function.

The results of both short-term and long-term exposure experiments conducted in the Beaufort Sea are reported and analyzed in Section IV of this report. There were, however, several basic differences observed in long-term effects of crude oil between the Elson Lagoon and the Kasitsna Bay studies that need to be stressed. The long-term effects of crude oil took much longer to be expressed in Beaufort Sea sediments than in Kasitsna Bay sediments. Reduced microbial activity was observed in sediments exposed to crude oil for only 5 weeks or less in Kasitsna Bay. It took up to a year before Elson Lagoon sediments showed the same shift. The same was true with percent respiration changes. From this, it seems quite likely that the effects of crude oil should last longer in Beaufort Sea sediments than in sediments from Cook Inlet.

In the Kasitsna Bay study, we found that the rate of nitrogen fixation can be depressed by 50-95% in oiled sediments. This change required only a few days exposure before measurable changes could be found. We have not observed a consistent reduction in nitrogen fixation rates in oiled Elson Lagoon sediments. Haines et al. (1980) have reported that denitrification rates are depressed in oiled Elson Lagoon sediments. It appears from their data, that the step affected involves the oxidation of ammonium ion to nitrite. This observation has serious implications relative to the overall impact of crude oil in Beaufort Sea sediments since nitrification is required to convert fix nitrogen in the form of ammonia to nitrate.

VI. Needs for further study

We feel that most of the cruise data (offshore) that is required for crude oil impact assessment has been collected. This is not true of the inshore areas, especially inshore environments near the major rivers and landward of the barrier islands. We feel that there is a very definite need for intensive study of microbial processes in the inshore sediments; especially those functions that relate to primary and secondary productivity. This would include both seasonal field studies and long-term effects studies. We have collected some of these data during the Elson Lagoon study; however, this study has not been comprehensive enough in terms of numbers of samples and geographical representation. We feel that a study of areas adjacent to one or two major rivers along the North Slope would be essential in defining the dynamics of microbial function and the potential effects of crude oil on those essential processes.

There are also some preliminary studies which could be conducted in areas that have already been impacted by crude oil; i.e. the oiled plots used by Dr. Dave Mason (RU #356) during the study "Environmental assessment of selected habitats in the Beaufort and Chukchi Sea littoral systems". Such a study would increase our knowledge of long-term crude oil effects in at least two diverse locations. These data would provide a good basis of comparison with what we have observed in Elson Lagoon and Kasitsna Bay.

One issue that has not been addressed at all in the Beaufort Sea is the effects of drilling muds on benthic microbial function. There are a number of components of drilling mud which could adversely affect a wide range of microbial functions and one component (paraformaldehyde) which would stop all microbial activities; that is why it is placed in drilling muds. A study of drilling mud effects could be conducted using the same methodologies that we are using to assess crude oil and dispersant effects.

NORTON SOUND

Section III

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development.

A. Objectives.

Our main objective during the July 1979 Norton Sound cruise was to determine if the microorganisms in areas near the gas vents in Norton Sound showed altered activity which would indicate the presence of petroleum hydrocarbons. There had been a study conducted in this area by Cline and Holmes (1977) which indicated there might be a natural seep of petroleum hydrocarbons in the Norton Sound, south of Nome, AK. We were to coordinate our research efforts with those of Drs. Cline, Kaplan, Feeley, and Atlas. In addition, we were to conduct measurements of relative microbial activity in both water and sediment samples and measure rates of nitrogen fixation in this region. There had not been any previous studies of microbial function in Norton Sound, thus we were to provide information to help fill a significant data gap that had existed in this region. It was important to obtain these data since it was assumed that a significant terrestrial nutrient imput was coming into this area from the Yukon River.

B. Conclusions and implications.

1. Data collected by Dr. Atlas, Dr. Cline, Dr. Kaplan, and ourselves indicate that the gas seep south of Nome that had previously been reported by Cline and Holmes (1977) did not contain significant levels of petroleum hydrocarbons. In fact, Kaplan, who was studying heavy hydrocarbons in the sediments of Norton Sound, did not find

any evidence for the presence of petroleum hydrocarbons in this whole area. Our data did not show any consistent patterns of reduced acute crude oil effects in either the waters or the sediments of this area (reduced acute effects in this application would indicate prior exposure to petroleum hydrocarbons).

2. Patterns of relative microbial activity and percent respiration show that there is a significant input of terrestrial carbon coming from rivers flowing into Norton Sound. The same patterns were observed here as we have seen in both Cook Inlet and Beaufort Sea where there is a significant freshwater input from a large landmass drainage. Similar patterns were also seen in the relative microbial activities in the sediments.

3. During the October 1980 Norton Sound Synthesis Meeting held in Anchorage, there was a large amount of data presented by the benthic ecologists and mammologists (Feder and Burns) which indicate that there is a substantial population of organisms which are totally dependent on the detrital food chain for food. This is particularly true of the organisms that overwinter in this area. If crude oil settles into the sediments of this region as a result of a spill, we would predict that much of this food source would no longer be available to these organisms.

4. The sediments along the north coast of Norton Sound showed the highest nitrogen fixation rates. If crude oil became incorporated into sediments in this area, the most significant rates of nitrogen fixation in Norton Sound would be affected.

II. Study area

During this cruise, we collected 62 water and 35 sediment samples at the locations illustrated in Fig. 29.

III. Methods

The methods used during this cruise were essentially the same as those described in Section I of this report.

IV. Results

A. Relative microbial activity

1. The mean values for relative microbial activity using both glucose and glutamic acid in the waters and sediments of Norton Sound were generally higher than that observed in the Beaufort Sea during two cruises (Table 16). These differences are not considered significant, however, because most of the locations sampled in the Norton Sound were in the unusually active region associated with Yukon River terrestrial carbon input. If we compare the relative microbial activity in Norton Sound with similar areas in the Beaufort Sea where there is terrestrial carbon input from major rivers, there is very little difference. The same general statement can be made when comparing Norton Sound data with that collected in the Cook Inlet (see Table 2, Section I).

2. Geographical distribution.

In the Norton Sound, we have seen the same patterns of relative microbial activity and respiration percentages that we observed in areas of major terrestrial carbon input in both Cook Inlet (Section I) and in the Beaufort Sea (Section II). The patterns of surface water salinity (Fig. 30) show that there are two major water masses in the area. One of these water masses has characteristics similar

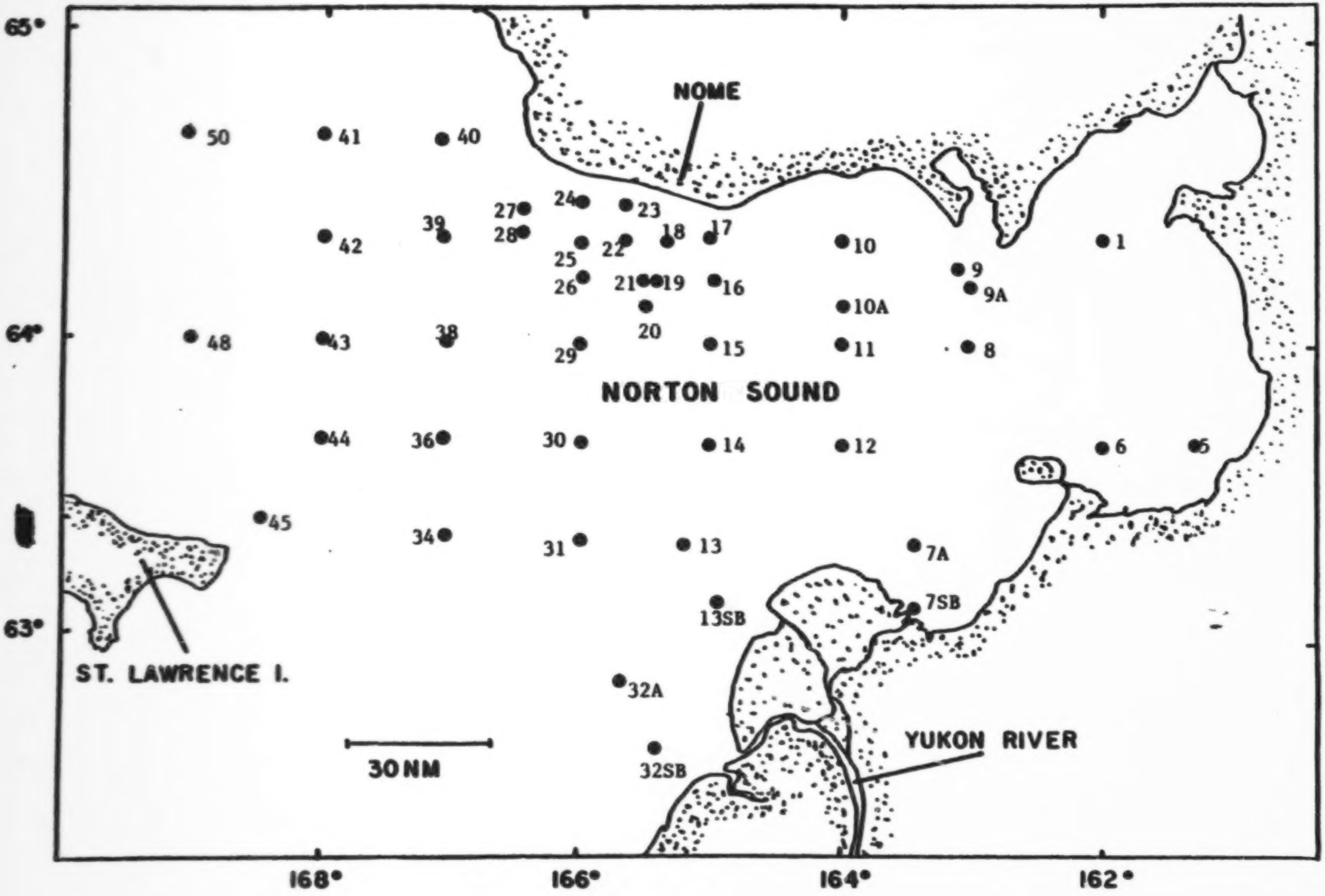
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Figure 29. Stations sampled during the Norton Sound cruise in July, 1979.

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Table 16. Comparison of microbial uptake rates for Norton Sound and Beaufort Sea summer water and sediment samples.

Location		Water				Sediment			
		Glucose[1]		Glutamate[1]		Glucose[2]		Glutamate[2]	
		mean	range	mean	range	mean	range	mean	range
Norton Sound	Jul 1979	12	1.1-110	19	1.5-182	28	0.1-154	127	3-1063
Beaufort Sea	Aug 1976	5	1-13	8	0.5- 24	4	1-15	80	20-180
Beaufort Sea	Aug 1978	7	1-44	14	1-14	9	1-24	96	7-262

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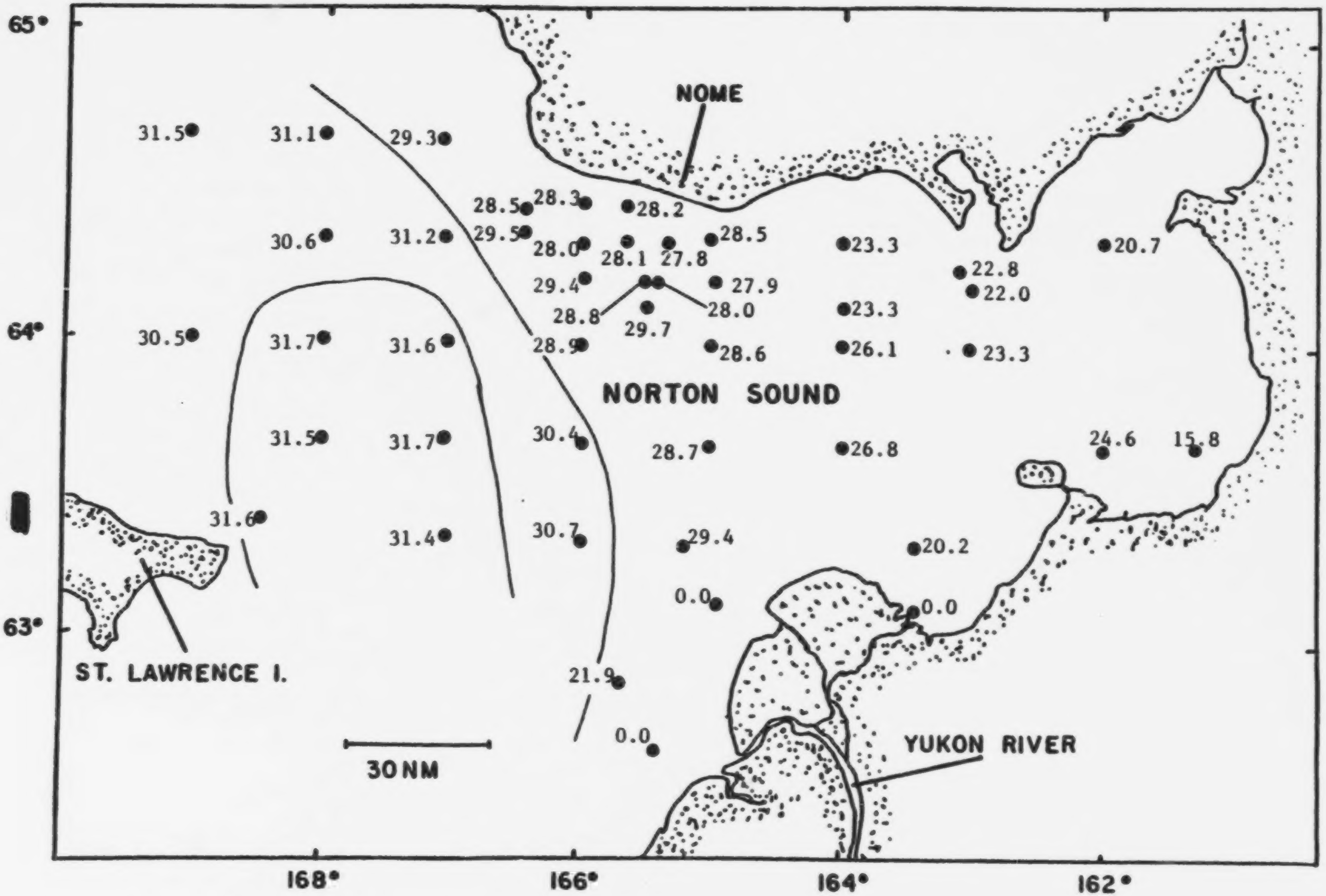
1 - ng/l/hr

2 - ng/g dry wt/hr



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29.4
27.9
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29.7
28.0
23.3
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28.9
28.6
26.1
23.3
31.5
31.7
30.4
28.7
26.8
24.6
15.8
31.6
31.4
30.7
29.4
20.2
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0.0
21.9
0.0

Figure 30. Surface water salinity as parts per thousand during the July 1979 cruise.

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to open ocean water that we have studied in the past. This water mass was found in a region which was within a 75 mile radius from the eastern tip of St. Lawrence Island. These waters showed high salinity, low relative levels of microbial activity and high respiration percentages in the microbial populations (Figs. 31 and 32). The waters analyzed at the other locations showed the reverse pattern. The lower salinities and higher relative levels of microbial activity show the impact of the freshwater input from the Yukon River (a major feature of the Norton Sound). A statistical analysis of the relationship between salinity and relative microbial activity indicates that there is an inverse relationship that is significant at the $p<0.0005$ level. The highest levels of microbial activity were observed in freshwater from the Yukon River.

If the water samples that are associated with the high salinity to the west (group A) and the water associated with the low salinity (group B) are compared, the differences become apparent. The mean rate of glucose uptake in group A was 1/2 that of group B but the significance of this difference for 47 samples was only $p = 0.068$. If the same comparison is made with the respiration percentages measured at the same time, the mean value for group A was 38% and the mean value for group B was 24%; a highly significant difference with a p - level = 0.000004.

In this region, there are also two water masses which form during the summer months in Norton Sound. A colder and more saline layer is located a few meters from the bottom and another layer on top of this extends to the surface. We found the mean rate of glucose uptake (relative microbial activity) to be twice as high in the bottom waters as that observed in the overlaying waters. This

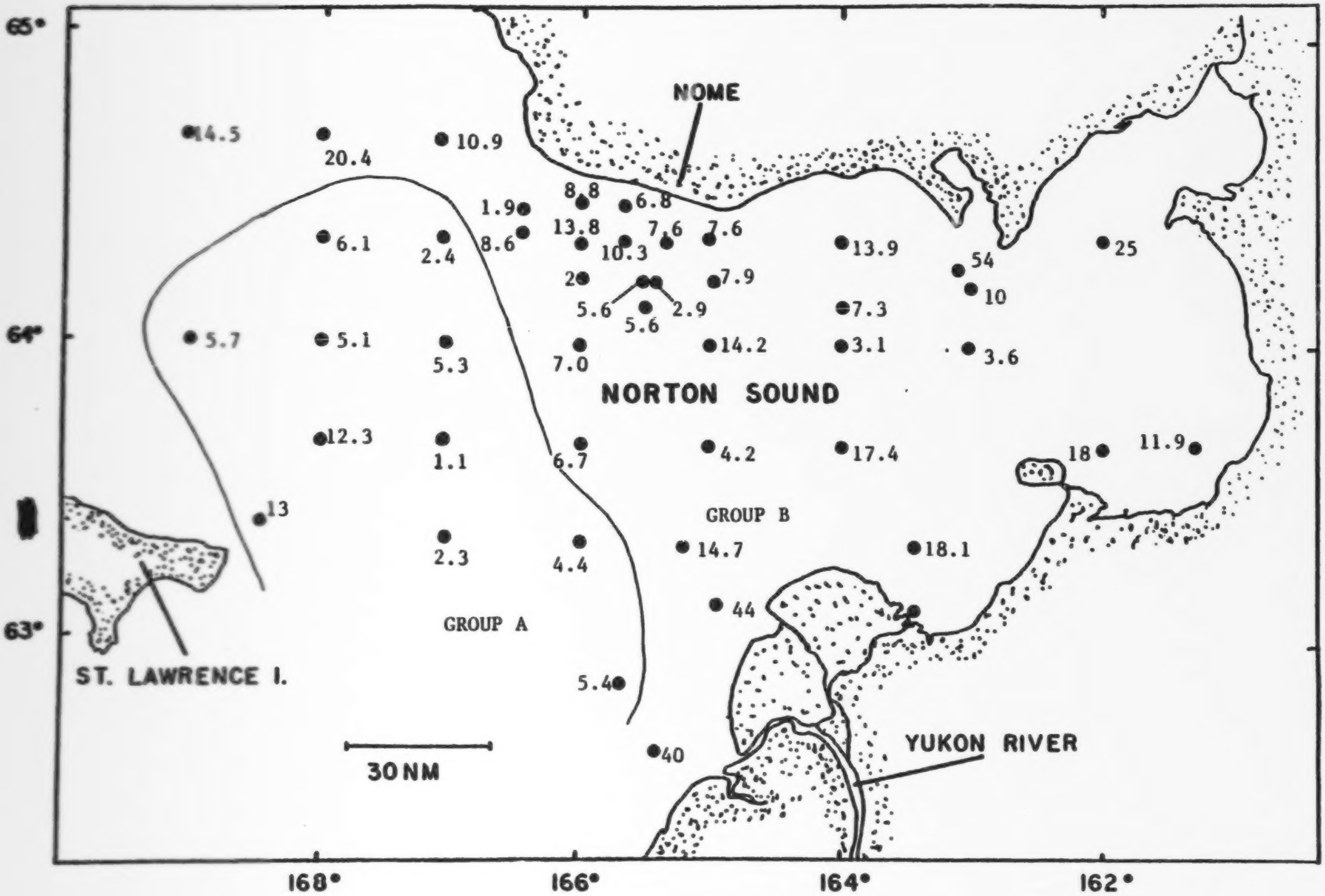
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Figure 31. Glucose uptake in ng/l/h, in surface water samples during the July 1979 cruise.

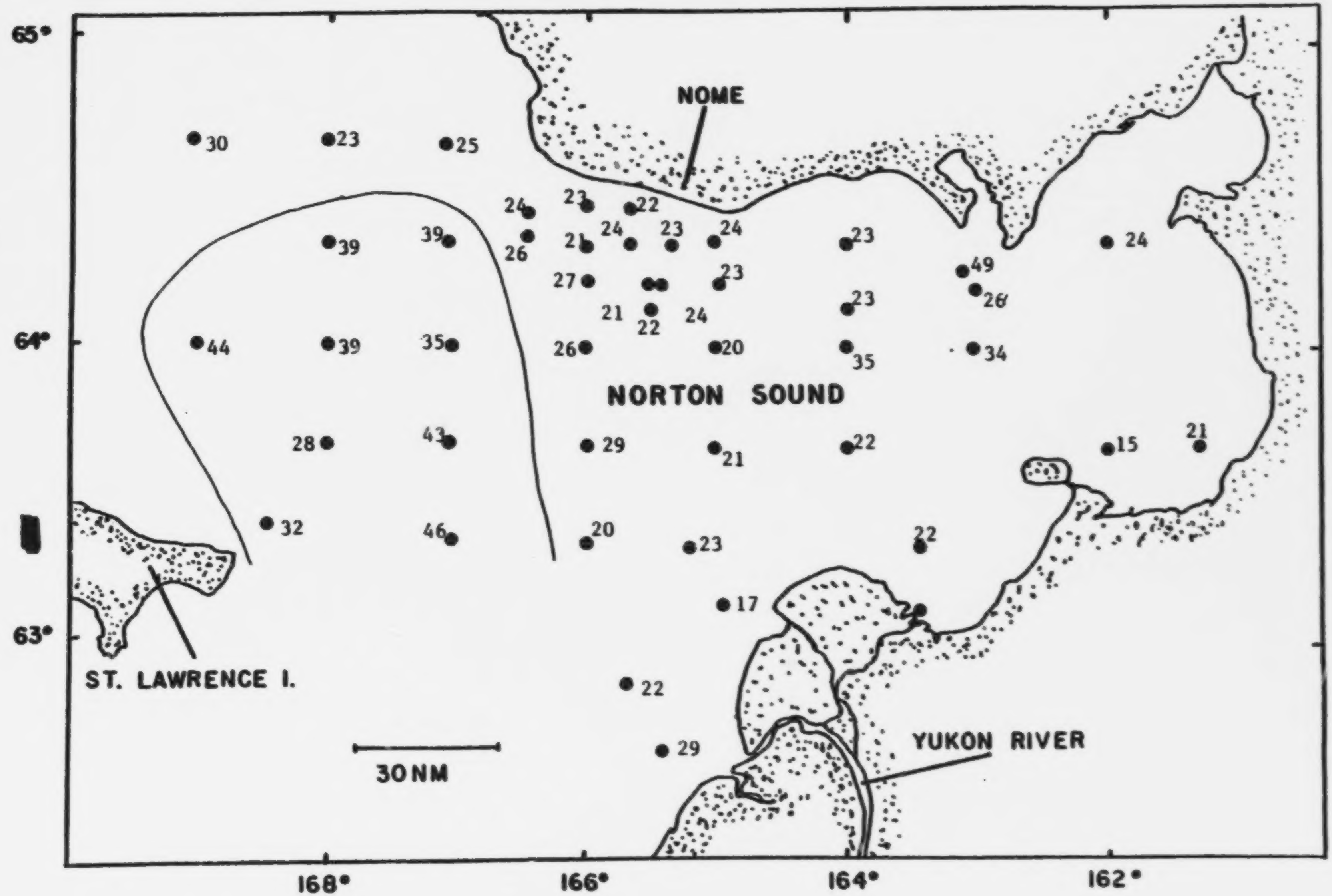

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Figure 32. Glucose percent respiration in surface waters during the July 1979 cruise.

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comparison was made at 14 locations and the significance of this difference was at the p = 0.02 level.

There were also geographical trends found in glucose uptake rates in the sediments of Norton Sound (Fig. 33). With the exception of the sample collected at station 41, the highest rates were observed in sediments collected in the fine-grained sediments in eastern Norton Sound.

B. Nitrogen fixation in Norton Sound sediments.

The mean nitrogen fixation rate observed during this cruise was approximately equal to that observed in the Beaufort Sea, but it was significantly lower (roughly 1/2 the rate) than that observed in the Shelikof Strait (see Table 4 in Section I). The highest rates of nitrogen fixation were observed along the north coast (Fig. 34).

C. Crude oil effects studies.

During this cruise, we measured the acute effect of crude oil on the uptake of glucose in both water and sediment samples (Figs. 35 and 36). The percent reduction in glucose uptake was determined by comparing the uptake rates of control samples with those exposed to crude oil. No significant geographical trends were noted.

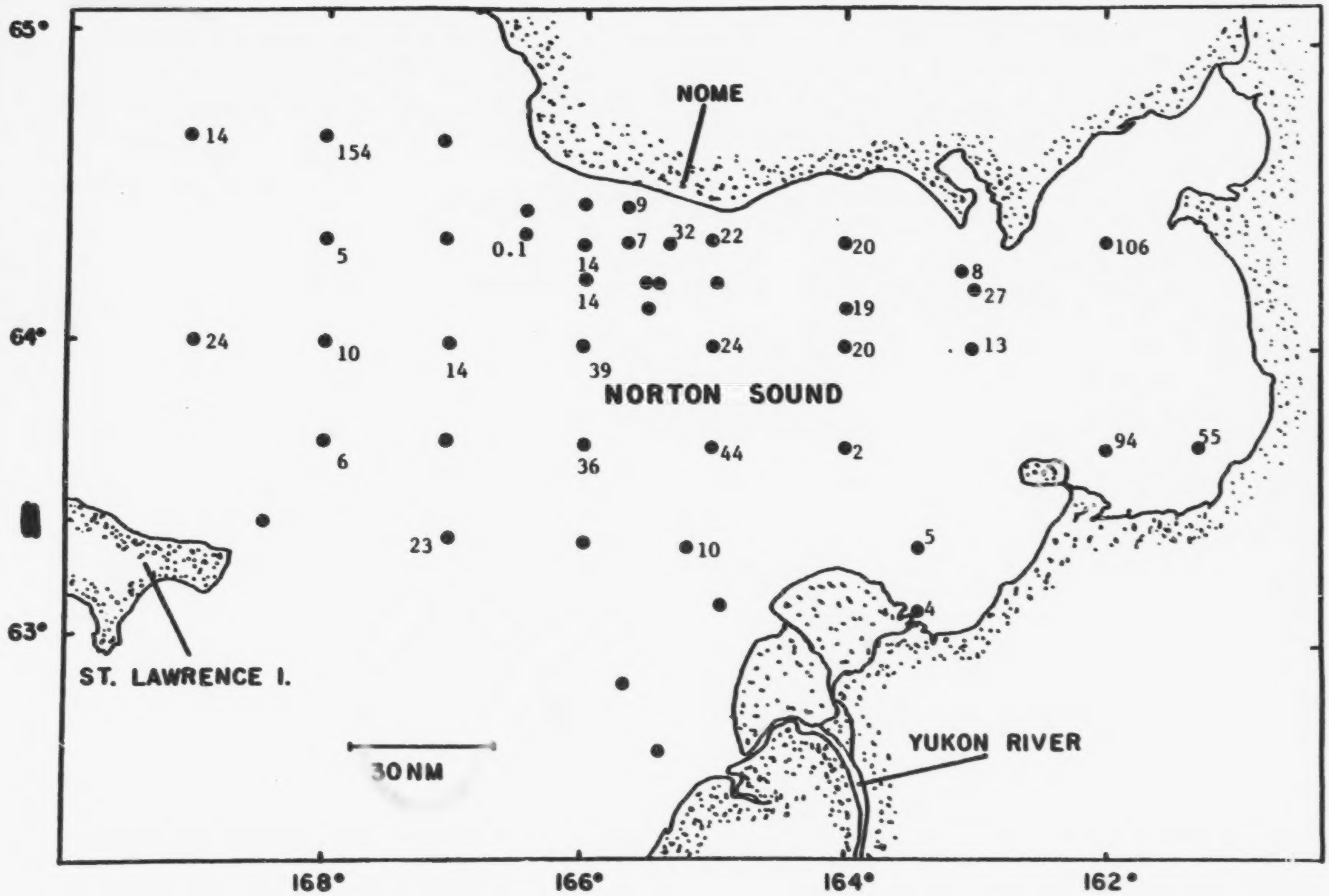
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Figure 33. Glucose uptake in sediments, expressed as ng/g dry wt./h., during the July 1979 cruise.

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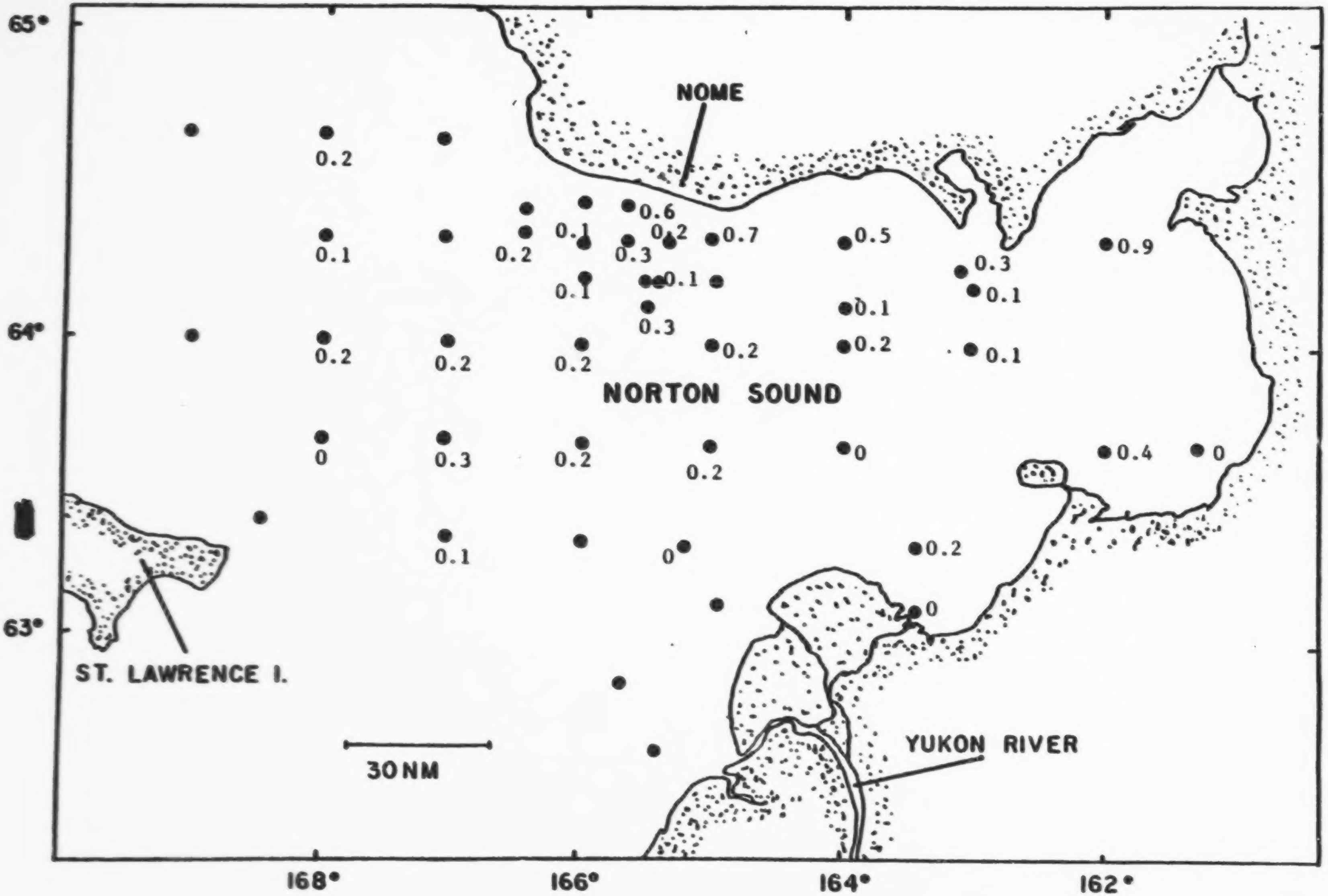

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Figure 34. Nitrogen fixation rates in sediments during the July 1979 cruise. The units are ng N_2 fixed/g dry wt./h.

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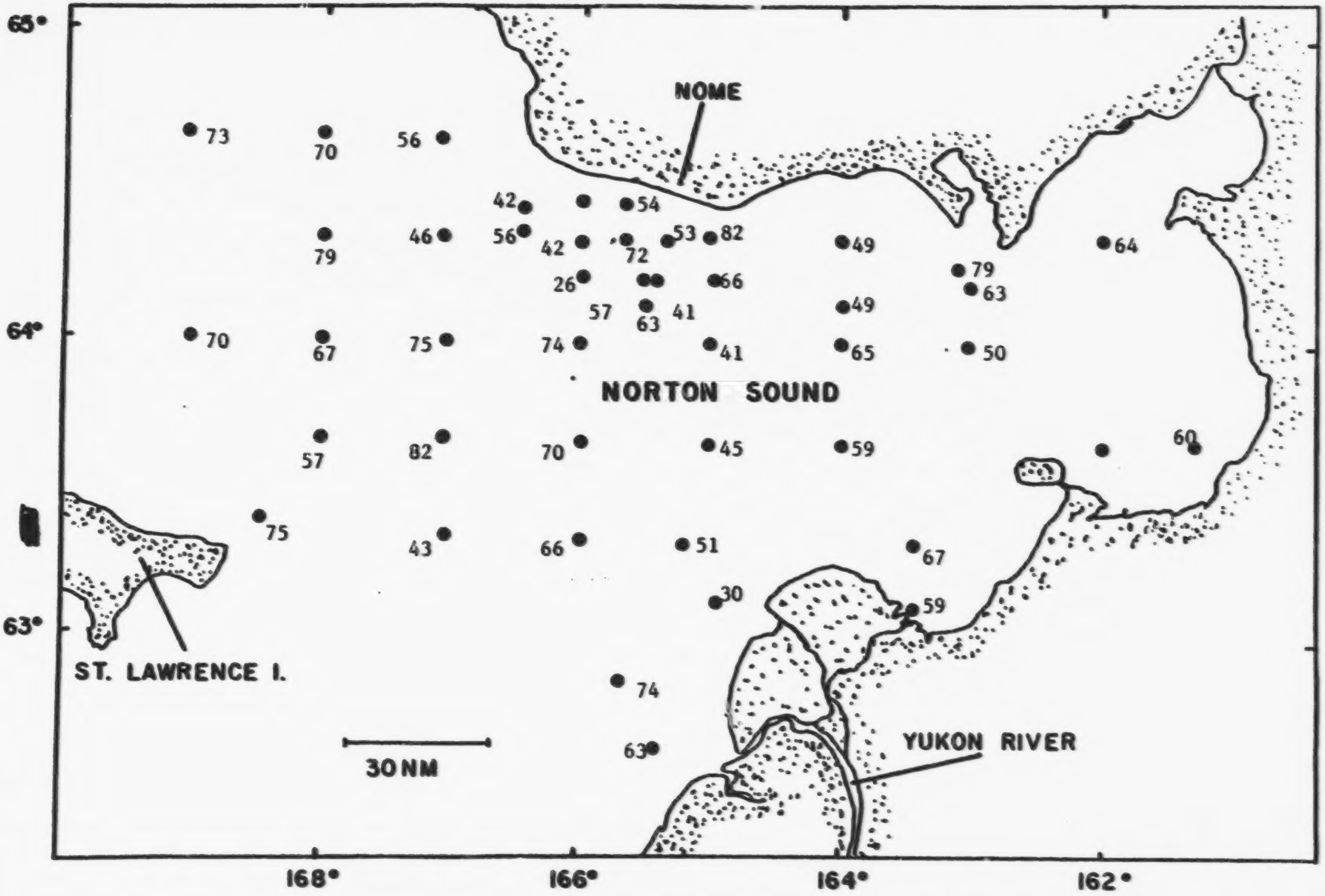

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Figure 35. Percent reduction in glucose uptake in surface water samples exposed to crude oil. July 1979 cruise.

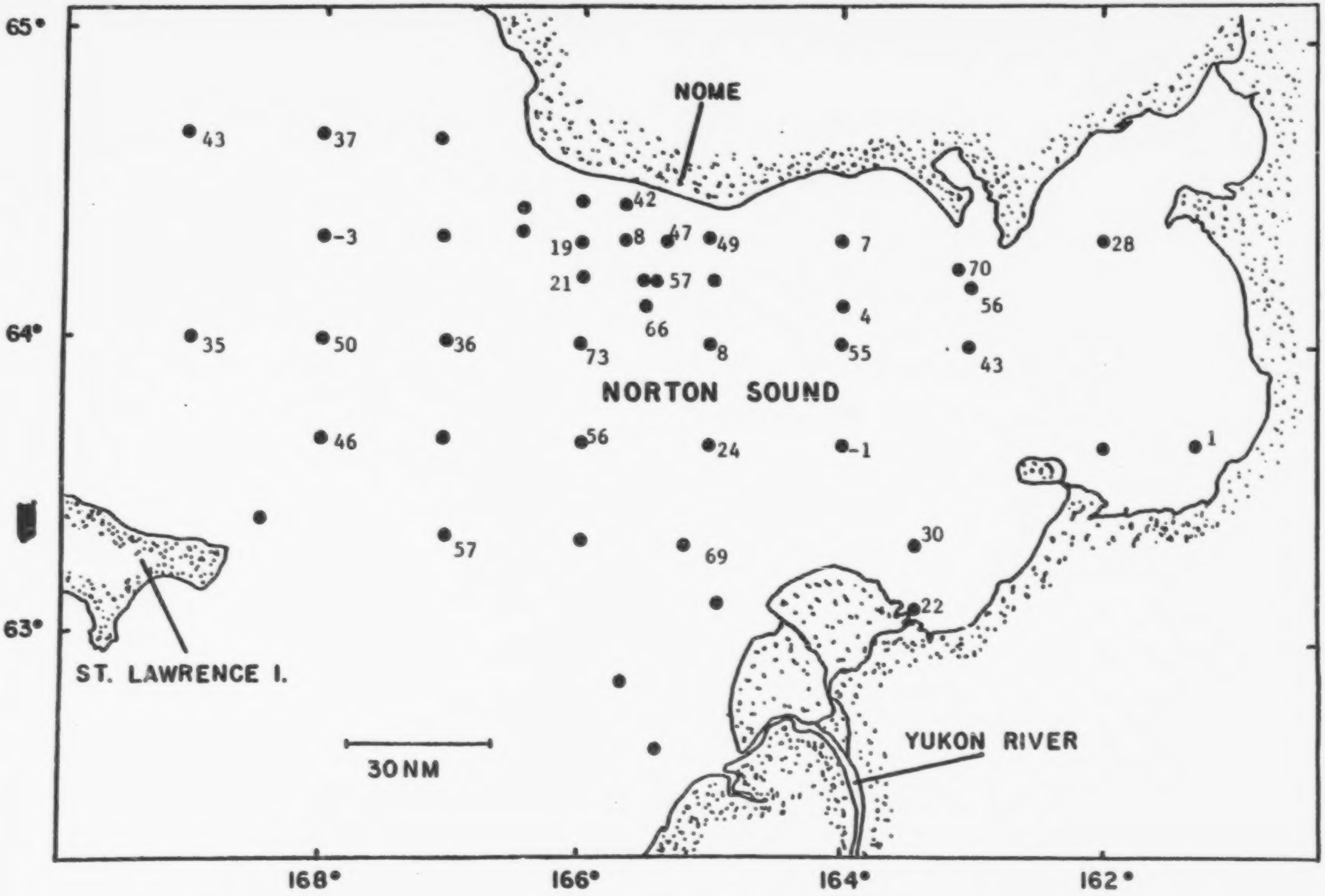
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Figure 36. Percent reduction in glucose uptake in sediments exposed to crude oil. July 1979 cruise.

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V. Discussion

The patterns of relative microbial activity, respiration percentages, and nitrogen fixation rates all suggest that there is a significant input of terrestrial carbon into Norton Sound. The relatively high microbial activity found in Norton Sound surface waters are an indication of this carbon input. The low respiration percentages in these waters suggest that the microbial population is actively growing because proportionately more of the carbon that is being utilized by the microorganisms is being incorporated into biomass than is respired as CO_2.

If the natural rates of nitrogen fixation are compared with denitrification rates (Table 5, Section I), it can be seen that the denitrification rates are much higher. It is felt that this condition reflects the input of exogenous organic carbon into the system. The high rates of microbial activity in the center and eastern end of Norton Sound could also reflect the input of exogenous carbon. In the Norton Sound, the most likely source of the organic carbon would be the Yukon River; although local input from the Yukon delta and the other rivers present may also contribute significant quantities of carbon as well.

The high microbial activity in the bottom waters of Norton Sound probably reflect the release of organic carbon from the sediments or a suspension of particles from the sediments. This is another case in which a distinct water mass can be characterized by measuring microbial function.

R. Feely has observed that the regions where most of the fine grained sediments from the Yukon River settle within Norton Sound are the same areas where we observed the highest relative microbial activity (see his final report). This suggests that there is a significant input of detrital material from the Yukon River which is actively being utilized by benthic microorganisms. H. Feder has observed that a large portion of benthic organisms found in the Norton Sound are detrital feeders (final report). During the October 1980 Norton Sound synthesis meeting held in Ancorhage, it was also mentioned by J. Burns that many of the mammals that come into Norton Sound feed on the detrital feeding benthic organisms found there. In addition, it was suggested that juvenile shrimp, crab and salmon also depend heavily on detritus as their main food source. In addition, all organisms that overwinter in the Norton Sound would depend exclusively on detritus (bacterial biomass) for food during the winter months since there is essentially no primary productivity taking place at that time. These observations suggest, that in Norton Sound, organisms at all trophic levels are dependent on the detrital food chain. As we will outline in Section IV of this report, the fact that most of the carbon coming into this system is probably terrestrial, suggests that most of the carbon must be cycled through bacterial biomass before it can be utilized by higher trophic levels.

As we have also reported in Section IV, there is strong evidence that the detrital food chain is very sensitive to perturbation by crude oil. We therefore suggest that if crude oil does get into the sediments of Norton Sound, there will be a reduction in the overall productivity of the region for an extended period of time.

One of the basic assumptions on which this cruise was based was that there was a petroleum seep in the Norton Sound. The methods that we used to determine chronic exposure of microbial populations to petroleum hydrocarbons showed no consistent patterns which would indicate the chronic input of petroleum hydrocarbons. Using a different methodology, Dr. Atlas also came to the conclusion that there were no chronic inputs of petroleum hydrocarbons. The recent studies conducted by the hydrocarbon chemists also support this conclusion.

VI. Needs for future research

The data collected during this cruise should be considered as a small part of a much larger study that needs to be conducted in the Bering Sea. At the present, there is very little known about the microbial processes in the Bering Sea and how they related to the overall ecology of the system.

This is a very important region for a number of reasons. First, there are a large number of proposed oil-lease tracts in the Bering Sea; several of which have high potential for the production of significant amounts of petroleum hydrocarbons. Second, this area is a major fishery - both bottom fish and crab - which is supported by a detrital food chain. At this time, we do not know what impact petroleum production and transportation will have on critical microbial processes in this system. Third, the Bering Sea represents a transition between the Beaufort Sea and Cook Inlet. We currently have data that crude oil affects various microbial processes in different ways when comparing long-term effects in Beaufort Sea and Cook Inlet sediments.

At this time, the St. Georges Basin in the southern Bering Sea would appear to be the region which should be given the highest priority. It has been concluded by the investigators involved in the PROBES study in this region, that the benthic detrital food chain is one of the most important features of this highly productive region (Iverson et. al, 1979).

Although it is now knows that bacterial biomass is the key component of detrital food chains in general, no direct observations of microbial function in this region have been made during the PROBES work and very few observations have been made during the course of the OCSEAP work in this region. Because of the potential importance of mcirobial function to the productivity in this area and the potential impact of crude oil on these processes in the case of a major spill, we recommend that the relative importance of key microbial processes be documented in the sediments and water column of the St. Georges Basin.

Although, at this point, we could hypothesize that these sediments may respond to the impact of crude oil in a way similar to Kasitsna Bay sediments; we recommend that both comprehensive cruise data and oil effects data be collected in the Bering Sea to determine the specific magnitude of such an impact.

EFFECTS STUDIES

Section IV

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development.

A. Objectives.

Our main objectives in these studies were to determine what effects crude oil and the crude oil dispersant Corexit 9527 have on major microbial functions in the waters and sediments of the Beaufort Sea, Norton Sound, and Cook Inlet. Due to time and technical constraints, we had to restrict our studies to short-term effects during the cruises in these areas. During these cruises, however, we were able to collect baseline data which would enable us to evaluate the potential impact of crude oil perturbations in various regions.

Two major studies were initiated to define the long-term effects of crude oil on microbial activities. One of these studies was initiated in Elson Lagoon (Beaufort Sea) in July 1977 and the other was initiated in February 1979 at Kasitsna Bay (Cook Inlet). These studies were designed to evaluate the long-term effects of crude oil and Corexit on relative microbial activity, nitrogen fixation rates, denitrification rates and respiration percentages.

B. Conclusion and implications.

1. We have found a broad range of both short-term (less than 24 h) and long-term (up to 2 years) effects of crude oil on major microbial functions.

2. Both crude oil and Corexit had an adverse (short-term) effect on relative microbial activity in water and sediments. When both of these were added in combination, the effects were even more

dramatic. The net effect of these perturbations was to place the microorganisms under stress which would undoubtedly reduce the species diversity of the organisms present over a period of time. When pelagic microorganisms were exposed to Corexit or crude oil for longer periods of time (4 to 5 days), the initial depression in microbial activity was essentially eliminated. Although there are problems in interpreting the results of this type of experiment, it did appear that Corexit or fresh crude oil did not suppress growth of those heterotrophic microorganisms that could utilize and mineralize glucose. From these observations, it would appear that at least this function in marine waters is not reduced after longer exposure to these pollutants.

3. Long-term exposure of marine sediments to crude oil altered a number of important functions. These included reduced microbial activity, increased respiration percentages, decreased bacterial biomass, decreased total biomass, reduced metabolic activity by benthic organisms, decreased nitrogen fixation rates, decreased denitrification rates, increased production of CO_2 and methane, increased sediment surface acidity and decreased surface redox potentials (reduced O_2 levels), decreased infaunal burrowing activities and increased accumulation of detrital particles on the sediment surface.

In addition, the activities of various enzymes were also altered by the presence of crude oil. Arlysulfatase, phosphatase, cellulase, laminarinase and chitobiase activities were depressed in the presence of fresh crude oil while amylase and alginase activities were increased.

4. The implications of these changes are described in detail under "Discussion" in this section. The major implications are that exposure of sediment to crude oil can act to reduce both primary and secondary productivity of the whole system over an extended period of time (years). A minimum reduction in overall productivity of 25% would be expected in areas where sediments contained 1 ppt fresh crude oil or more. In addition, the chemical composition of the sediments surface is altered so that normal recruitment of benthic organisms into the impacted area will also be altered for an extended period of time. This condition could well extend past the time when the direct toxic effects of the crude oil are no longer a factor in killing, injuring or repelling the organisms.

5. At least two important reactions in the nitrogen cycle are adversely affected by the presence of crude oil; nitrogen fixation and denitrification. Nitrogen fixation appears to be particularly susceptible to the effects of crude oil and as such would be a very important variable to measure in environmental impact studies of oil spills.

Such studies should also include measurements of relative microbial activity and respiration percentages. In addition, concentration determinations should be made for compounds which are associated with anaerobic fermentation; i.e. H_2S, methane, and ammonia. Measurements of oxygen concentrations in interstitial and interface waters would also be helpful.

6. We feel that the altered functions that we have observed would occur under actual spill conditions.

We have conducted studies in which the effects of various concentrations of fresh crude oil on benthic microbial processes have been observed. The results of these studies indicate that many of the changes that we have observed at the relatively high concentration of 50 ppt also occur at 1 ppt and even at concentrations as low as 0.1 ppt. Unfortunately, the types of measurements that we are conducting in our studies have rarely been conducted in studies of actual oil spills; however, where these observations have been made, they correlate well with our findings.

7. We have concluded that every effort should be made during the planning of crude oil production and transport to reduce the risk of incorporating crude oil into marine sediments. This would also apply to procedures used to clean up an oil spill as well. This could possibly lead to some difficult choices during the control of a crude oil spill. It can no longer be assumed that one of the main objectives of control is to remove the slick from the surface of the water. If this is done by driving the oil into the water column and then into the sediments, the risk of killing birds and mammals will have to be weighed against the possible reduction in the food supply for all organisms present for many years.

8. Of the areas that we have studied, the inshore waters of the Beaufort Sea and Kachemak Bay in Cook Inlet would appear to be the most vulnerable. We feel that the Beaufort Sea is particularly vulnerable since it appears that the effects will last longer there than in other areas we have studied; i.e. Cook Inlet. Also, this area may be more dependent than other areas on bacterial regeneration of the inorganic nutrients required for the spring phytoplankton

bloom. In Kachemak Bay, the major carbon input is thought to come from macrophytes and land plants. Most of this material must be cycled through the detrital food chain and is thus susceptible to crude oil perturbation.

One area that we have not studied that could be severely impacted by crude oil perturbation is the St. Georges Basin in the southern Bering Sea. There is evidence that suggests that the highly productive fisheries in the region is dependent to a large degree on the detrital food chain for its food.

II. Study areas.

Samples for the short-term effects studies conducted during cruises were collected at the locations shown in previous sections. The long-term crude oil effects study in Elson Lagoon was conducted at a location approximately half way between stations 2 and 3 shown in Fig. 16 of Section II. The sample used in the Kasitsna Bay study was collected at the location in Kasitsna Bay shown in Fig. 37.

III. Methods

Except for the procedures described below, the techniques used in these studies were the same as those described in Section I and II of this report.

A. Sample collection and manipulation. Sediment was collected from the bottom of Kasitsna Bay (35 - 50 m) using a pipe dredge. Combined dredgings were mixed in 120 liter plastic containers and treated with various concentrations of crude oil as listed below. Subsamples of the treatments were placed in a plexiglass trays (30.5 x 30.5 x 10.2 cm) and positioned on the bottom of Kasitsna Bay in 20 m of water by SCUBA divers. The trays were covered with

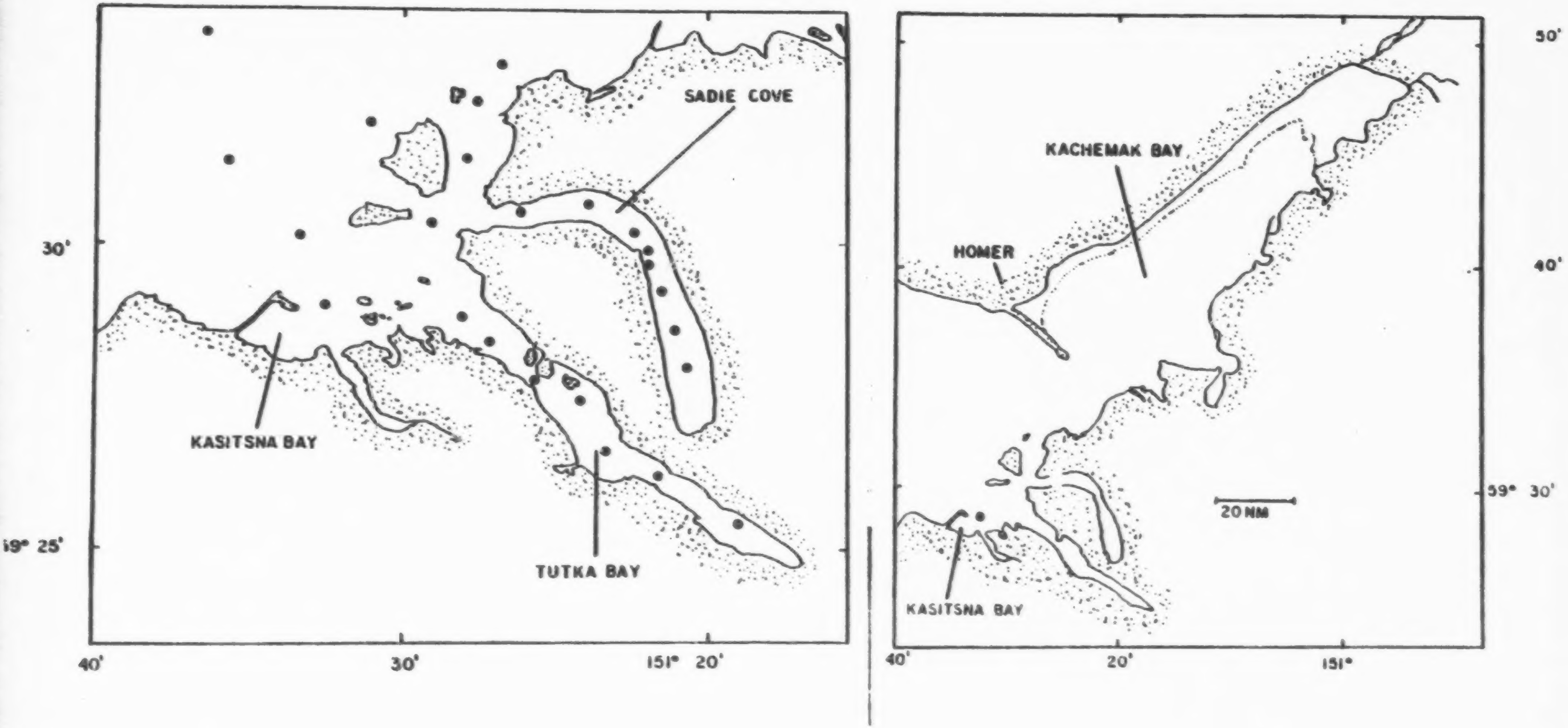
SADIE COVE
KASITSNA BAY
TUTKA BAY
30'
59° 25'
40'
30'
151° 20'
KACHEMAK BAY
HOMER
KASITSNA BAY
20 NM
50'
40'
59° 30'
40'
20'
151°

Figure 37. Location of sampling sites in the Kasitsna Bay area.

plexiglass lids to reduce sediment washout during raising and lowering. Since the analysis of the trays took approximately two days, the trays were kept in a trough through which fresh seawater was circulated at a rate of eight liters per minute. This kept the sediments at in situ temperature and insured the replenishment of dissolved gases. Subsamples for assays were removed from the trays using a 50 ml plastic syringe with the end of the barrel removed. Five to seven of the resulting cores were combined in a 500 ml plastic jar and then vigorously shaken to form a homogenous sediment slurry.

1. Initial experimental treatments consisted of preparing triplicate trays (8 liter sediment volume) with untreated sediment and sediment mixed with Cook Inlet Crude to give a final concentration of 50 ppt (v/v). To examine the effect of a dispersant, duplicate trays were prepared with and without crude oil and with either 50 or 500 ppm final concentrations of Corexit 9527 (Exxon).

Subsamples of the treated sediments used in the tray experiments were also placed in aquaria. Four aquaria were set up with one each containing non-treated, crude oil (50 ppt), crude oil and 50 ppm on Corexit 9527, and crude oil and 50 ppm Corexit (32 liter sediment volume). Ten cm high plexiglass dividers were placed across the bottom of the aquaria to make two sections which could be sampled separately. Fresh seawater was supplied at one end of each aquarium and removed at the other end by a constant-level siphon system which replaced the volume four to six times each hour. The seawater flow system at the laboratory consisted of a stainless steel pump located 125 m offshore. The only materials that the seawater came in contact with were PVC plastic, natural latex rubber tubing, nylon, and stainless steel.

2. Concentration series and oil overlay experiments using both fresh and weathered crude oil were replicated in either one liter plastic freezer containers or trays respectively. The weathered crude oil was prepared by floating fresh crude oil on seawater exposed to outdoor conditions for six weeks and then skimming off the residue. Fresh oil concentrations used included 0.01, 0.1, 0.5, 1.0, 5.0, 10 and 50 ppt. Weathered oil experiments were conducted using 1.0 and 50 ppt concentrations.

Fresh and weathered oil overlays were constructed by adding eight liters of homogenous sediment to duplicate trays and then overlaying with one liter (approximately 1 cm thickness) of sediment with either 50 ppt fresh or weathered crude oil. In August 1979, the two aquaria with Corexit treatments were terminated and replaced with a control and 50 ppt weathered crude overlay of the same proportions as the tray experiment.

3. Studies of sediments augmented with organic nutrients were established at several different intervals. Initial studies were performed in trays divided into four equal sections with plexiglass dividers. Each quadrant contained a different nutrient amendment: none; 5% (w/v) Cerophyl (dehydrated cereal grass leaves obtained from Cerophyl Laboratories, Kansas City, MO, USA); 5% unmodified wheat starch (Sigma); and 5% chitin (Sigma). Tray treatments

consisted of control, 50 ppt fresh crude oil, and crude oil with either 50 or 500 ppt Corexit.

Later experiments were run in 300 or 500 ml plastic jars with 2% (w/v) concentrations of cellulose, starch, chitin, raw seaweed (*Alaria* sp., dried and ground to pass a 0.5 mm sieve), and gelatin. All were run with and without fresh crude oil at 50 ppt.

4. Additional tray experiments were established at the head of Sadie Cove and Tutka Bay, and in Mud Bay behind the Homer Spit (Figure 37), and at Elson Lagoon near Point Barrow using sediments from those locations. Sadie Cove and Tutka Bay duplicate trays were treated with either 50 ppt fresh or weathered crude oil in addition to controls. A nutrient amended tray set with control and 50 ppt fresh crude oil was also placed at Sadie Cove. Mud Bay treatments included duplicate trays with either 50 ppt fresh or 1 ppt weathered crude oil; and a set of nutrient amended jars with seaweed, alderleaves, and sawdust.

B. Procedures for assaying the effects of crude oil and Corexit on relative microbial activity.

1. The assays were conducted in essentially the same way as those described in Section I using the following isotopes:

14C-Glucose - 291-336 mCi/mM, 5-6 μg/L
14C-Glutamate 285 mCi/mM, 5 μg/L for water; 10 mCi/mM, 150 μg/L for sediment dilutions
14C-Acetate 57.8 mCi/mM, 5 μg/L
14C-Glycollate 35 mCi/mM, 11 μg/L.

2. The acute effects of crude oil and Corexit 9527 were measured by adding various combinations of 10 μl fresh crude oil and

15 or 50 ppm Corexit to replicate 10 ml portions of either seawater or sediment dilution. In assays where both oil and Corexit were used, the dispersant was added after the crude oil. The effect of low-level Corexit exposure on glucose uptake and respiration was determined for final dispersant concentrations 1, 5, 10, 15, and 20 ppm.

3. Intermediate term effects of Corexit on relative microbial activity were assayed by incubating seawater and sediment dilutions with 50 ppm Corexit for up to 14 hours at in situ temperature. At various intervals, replicate 10 ml portions were assayed for glucose uptake activity (1-2 hour incubation).

Interactions of various nitrogen and phosphorus nutrients with Corexit effects were examined by adding combinations of 50 mM glutamate, 5 or 500 μM NH_4Cl, and 0.5 μM Na_2HPO_4. The treatments were also challenged with 10 μl fresh crude oil and/or 50 ppm Corexit.

4. The water-soluble fraction of Cook Inlet Crude oil was prepared by slowly shaking 5 ml oil in one L filter-sterilized seawater at 10°C for 48 hours and then siphoning off the non-oiled subsurface water. The aqueous phase was then re-sterilized by filtration. Five ml of various dilutions of aqueous phase and sterile seawater was mixed with 5 ml of either fresh seawater or sediment dilution and assayed for glucose uptake in the normal manner.

C. Enzyme Assays

1. Arylsulfatase and phosphatase determinations were conducted using modifications of techniques originally described by Tabatabai and Bremner (1969, 1970). These assays are based on the enzymatic release of p-nitrophenol from the appropriate chromogenic substrate. To one ml of sediment slurry was added: one ml 30 ppt Rila Marine Mix - 0.05 M Tris buffer, pH 7.5; and one ml of either 0.006 M p-nitrophenylphosphate or 0.006 M-p-nitrophenylsulfate (Sigma) in buffer. Substrate and sediment blanks were run by omitting either the sediment slurry or the substrate. The reaction mixture was incubated at the original *in situ* temperature for one hour and then terminated with two ml 0.5 N NaOH and 0.5 ml of 0.5 M $CaCl_2$. The sample was then centrifuged and the optical density of the clear supernatant at 410 nm was measured to determine the amount of p-nitrophenol released. A calibration curve was prepared using dilutions of 10 μM per ml p-nitrophenol (Sigma). Samples with oil required two centrifugations to eliminate spurious turbidity due to oil droplets. Final results were calculated as μmoles p-nitrophenol released x gram dry weight^{-1} x hour^{-1}.

2. Chitobiase determinations were run identically to arylsulfatase and phosphatase except the substrate was 0.006 M p-nitrophenyl-N-acetyl-β-D-glucosaminide (Sigma) and the incubation time was four hours.

3. Polysaccharide hydrolase assays were based on the colorimetric determination of reducing sugars enzymatically released from appropriate substrates. To three ml of sediment slurry was

added three ml 30 ppt Rila Marine Mix - 0.05 M Tris buffer containing one of the following: 1% soluble starch, 1% carboxymethylcellulose, 0.5% laminarin or 0.5% sodium alginate. Three tenths ml of toluene was added to limit microbial metabolism during the assay. Substrate and sediment blanks were run by omitting either the sediment slurry or the substrate. The reaction mixture was incubated at the original in situ temperature for 24 hours and then centrifuged. Two ml of the resulting supernatant was added to two ml of DNSA reagent. (This reagent was prepared by dissolving one gram of 2,5-dinitrosalicylic acid (Sigma) in 20 ml 2 N NaOH, adding 30 ml distilled water followed by 30 grams of sodium potassium tartrate, and then bringing the solution to a final volume of 100 ml with distilled water). The DSNA - supernatant mix was developed by placing in a boiling water bath for 10 minutes and then cooling. The mixture was centrifuged to remove precipitants and the optical density at 540 nm measured to determine the reducing sugar (as glucose) content. A calibration curve was prepared using dilutions of one mg per ml glucose solution. Final results were calculated as μg glucose (or equivalent) released x gram dry weight^{-1} x hour^{-1},

D. Redox potential and pH measurements.

Oxidation-reduction potential (mV) and hydrogen ion concentration (pH) were measured at in situ temperature on the surface and in the bottom third of undisturbed samples using platinum redox and pH electrodes (Orion) respectively. The redox electrode was calibrated using two buffer solutions: 0.1 M $K_4Fe(CN)_6$ + 0.05 M $K_3Fe(CN)_6$ for

+192 mV and 0.01 M $K_4Fe(CN)_6$ + 0.05 M $K_3Fe(CN)_6$ + 0.36 M KF and for +258 mV.

E. Primary productivity rates.

Primary productivity was measured as the uptake of $^{14}C\text{-}HCO_3^-$ by the method of Strickland and Parsons (1972). Samples were incubated with 5 µCi bicarbonate (New England Nuclear) for three to six hours at water surface temperature and light levels. Alkalinity was determined as 810 X salinity (R. Horner, personal communication). Final results were calculated as mg carbon fixed x m^{-3} x $hour^{-1}$.

F. Denitrification.

Denitrification rates and potentials were measured by assaying N_2O released when a sample was incubated anaerobically with the normal endproduct formation (N_2) blocked by acetylene; a technique described by Balderston et al, 1976 and adapted to marine systems by Sørensen 1978. Our procedure consisted of placing ten ml of sediment slurry in 60 ml serum bottles which were sealed rubber closures and flushed with argon at ten cm^3 per second for one minute. Ten ml of the 50 ml headspace was replaced with acetylene. Denitrification rates were measured by incubating at in situ temperature for ten days and then terminating with one ml formalin. Denitrification potentials were measured by adding one ml 0.01 M KNO_3 and for 24 hours before termination. All samples were sealed with silicon rubber cement and returned to O.S.U. for gas analysis.

N_2O was quantified on a Hewlett Packard model 5840A gas chromatograph using an electron capture detector after separation at 50°C on a 6.1 m x 0.3 cm diameter stainless steel column packed with 50/80 mesh Popopak Q (Waters Associates). The carrier gas was a mix of

95% argon and 5% methane flowing at 30 cm^3 per minute. A unit per area calibration was prepared using 100 ppm N_2O (Alltech Associates). Final results were calculated as ng N_2 (as N_2O) released x gram dry $weight^{-1}$ x $hour^{-1}$.

IV. Results

A. Short-term effects of crude oil and/or Corexit 9527 (24 hrs or less).

1. Crude oil.

a. Effects of relative microbial activity.

We have studied the effects of Alaskan crude oil on 215 water and 162 sediment samples collected from three very different regions along the Alaskan coast. These are all areas which could potentially be impacted by crude oil as the result of crude oil production and transportation. One of the goals of this study was to determine if the presence of crude oil would alter microbial function in these diverse marine environments. It was found that in the 7 field studies where the effect of crude oil on uptake of labelled glucose by pelagic microorganisms was analyzed, there was a statistically significant difference between the treated and non-treated samples (Table 17). The range in the mean percent reduction values observed for glucose uptake rates was from 37 to 58%. The statistical significance of these differences ranged from $p<0.035$ to $p<0.00001$. In seven out of eight of the field studies where sediment samples were analyzed, there was also a statistically significant difference between glucose uptake rates in treated and non-treated sediments (Table 17). The mean percent reduction values observed in the sediment samples ranged from 14 to 36%. The differences in the mean values observed (when comparing different regions) are probably not significant since the range in values observed at one location (Kasitsna Bay) were comparable. Even though both pelagic and benthic microorganisms were affected by the presence of crude oil, the benthic microorganisms were affected to a lesser degree. The

Table 17. Percent reduction of glucose uptake rates in microbial populations exposed to crude oil.

Sample	$\bar{Y}$	SD	n	range	*p<
Water					
Beaufort Sea, September, 1978	52	20	40	-8 - 88	0.0001
Cook Inlet, April, 1978	45	30	32	-43 - 86	0.01
Cook Inlet, May, 1979	58	20	47	-22 - 92	0.0005
Norton Sound, July, 1979	57	15	60	18 - 82	0.0001
Kasitsna Bay, February, 1979	37	13	7	22 - 83	0.012
Kasitsna Bay, April, 1979	58	16	6	36 - 72	0.035
Kasitsna Bay, July, 1979	56	11	23	0 - 72	0.0001
Sediment					
Beaufort Sea, September, 1977	°35	18	20	0 - 65	0.002
Beaufort Sea, September, 1978	32	22	34	0 - 68	0.001
Cook Inlet, April, 1978	14	32	14	-73 - 35	NS
Cook Inlet, May, 1979	29	42	14	-52 - 78	0.048
Norton Sound, July, 1979	36	22	34	1 - 73	0.0001
Kasitsna Bay, February, 1979	25	14	12	0 - 49	0.01
Kasitsna Bay, April, 1979	16	13	11	-2 - 42	0.01
Kasitsna Bay, July, 1979	30	12	23	0 - 72	0.0001

° = CO_2 data only.
* = level of statistical significance between treated and non-treated samples.
NS = not statistically different at the $p<0.05$ level.

significance of that difference is not known; perhaps the benthic microorganisms are more consistently exposed to biogenic hydrocarbons.

We wanted to determine if a similar crude oil effect might be observed if an amino acid was used as a substrate in the heterotrophic potential method. During the 1978 Cook Inlet cruise, the same experiment was conducted on 35 water and 7 sediment samples using labelled glutamic acid. The mean percent reduction observed was 33 and 18 respectively. The difference in uptake rates between treated and nontreated water samples was significant at the $p<0.0003$ level but the difference in the sediment samples was not statistically significant. It can thus be said that, at least in pelagic microbial communities, the effect of crude oil on heterotrophic rates is not limited to glucose uptake and respiration.

Since the single concentration method (Griffiths et al., 1977) was used to measure changes in uptake and respiration rates in populations exposed to crude oil, it could be argued that the differences observed might have been caused by some component or components of the crude oil which are competing for the same transport mechanisms that are being used to take up both glucose and glutamic acid. In order to determine if this is the case, we elected to use the Wright and Hobbie (1966) technique for measuring uptake kinetics. By using this technique the maximum potential uptake rate (V_{max}), the turnover time required to utilize all of the naturally occurring substrate by the microbial population (T_t) and transport constant plus the natural substrate concentration (K_t+S_n) can be calculated. If some component of crude oil is being transported into the cells via the same mechanism as glucose, the V_{max} value should not change but the T_t and the K_tS_n values should increase. A study on the effects of crude oil on the kinetics of glucose uptake was conducted on 6 water samples collected in the Beaufort Sea (Table 18). The mean V_{max} value decreased from 14.6 in the non-treated samples to 3.7 µg per liter per hr in the treated samples. The mean T_t value increased from 177 to 492 hr and the K_t+S_n value remained unchanged. The differences observed in both the V_{max} and the T_t values were statistically significant. It thus appears that crude oil is acting as a metabolic inhibitor.

During the course of our studies we also measured the percent respiration. We did observe differences in the percent respiration between treated and non-treated samples; however, these differences were not consistent or statistically significant in most cases. The mean values for treated samples were usually slightly higher than those observed in non-treated samples. These data suggest that the microbial function most affected is substrate transport. If either biosynthetic or respiratory functions were consistently affected, there would be a significant change in the percent respiration values in treated samples.

In one of the studies, we measured the effects of both fresh and "weathered" crude oil and an aqueous extract of fresh crude oil on respiration rates in 20 sediment samples. The average percent reduction was 20, 21, and 23% respectively. It would thus appear that under these conditions the effects of these various treatments on glucose uptake were essentially the same.

In another experiment, we showed that various concentrations of fresh crude oil aqueous extract affected the incorporation of glucose into microbial biomass (Fig. 38). The highest concentration used in

Table 18. The effects of crude oil, Corexit 9527, and a combination of the two on the kinetics of glucose uptake in water samples collected during the summer, 1978 Beaufort Sea field studies.

	No Oil		Crude Oil		Corexit		Corexit + Oil	
	$\bar{x}$	S.D.	$\bar{x}$	S.D.	$\bar{x}$	S.D.	$\bar{x}$	S.D.
(1)V_{max}	*14.6	11.3	*3.7	1.8	°2.7	1.3	°1.5	.9
(2)T_t	$228	151	$492	263	°1145	505	°1162	332
(3)$K_t + S_n$	$2.4	1.0	$1.9	1.3	°3.4	2.2	°2.5	1.0

(1) Maximum potential uptake rate: mean and standard deviation in μg/liter/hour.

(2) Turnover time: mean and standard deviation in hours.

(3) Transport constant + natural substrate concentration: mean and standard deviation in μg/liter.

* 7 samples

° 3 samples

$ 6 samples

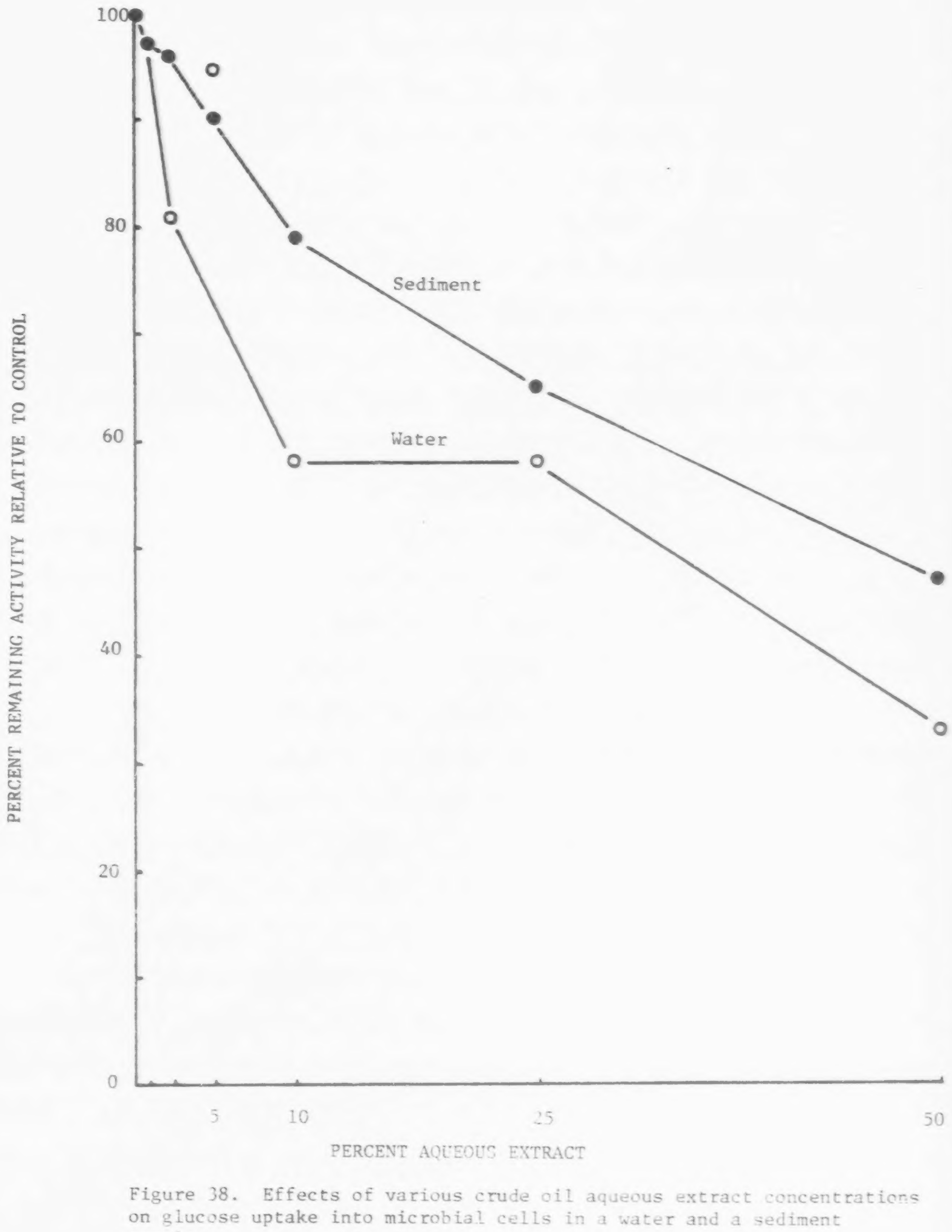

Figure 38. Effects of various crude oil aqueous extract concentrations on glucose uptake into microbial cells in a water and a sediment sample.

this experiment was 50% of that used in the studies reported above. This experiment clearly suggests that the soluble fraction of fresh crude oil adversely affects the incorporation of glucose into microbial biomass in both pelagic and benthic populations and that this effect is seen at concentrations much lower than that used in the field studies.

Although similar studies with "weathered" crude oil were not conducted; in this series of observations, we have observed that "weathered" crude oil has a similar effect on many microbial functions to that observed when sediments are exposed to fresh crude oil for one year. These observations suggest that the crude oil component that is inhibiting microbial biosynthesis may not be in the more volatile fraction.

In all of the above mentioned studies, the exposure time to crude oil was 12 h or less. We also conducted two studies in which water samples were exposed to fresh crude oil for longer periods of time. In one experiment, we used a water sample from the Beaufort Sea (Fig. 39) and in the other we used a water sample from Kasitsna Bay (Fig. 40). In both studies, the uptake rate for the substrates being tested decreased initially in the presence of crude oil but as the exposure time continued, the observed uptake rates in the oiled samples equalled and then surpassed the rate observed in the non-oiled samples. It must be cautioned that these experiments were carried out under very artificial conditions even though the incubation temperature was within 1°C of the *in situ* temperature. The large increase in activity observed in both cases is undoubtedly due to the well documented "bottle effect" which is always observed when marine water samples are confined within an incubation vessel. Even with this in mind, it appears that the populations are not only adjusting

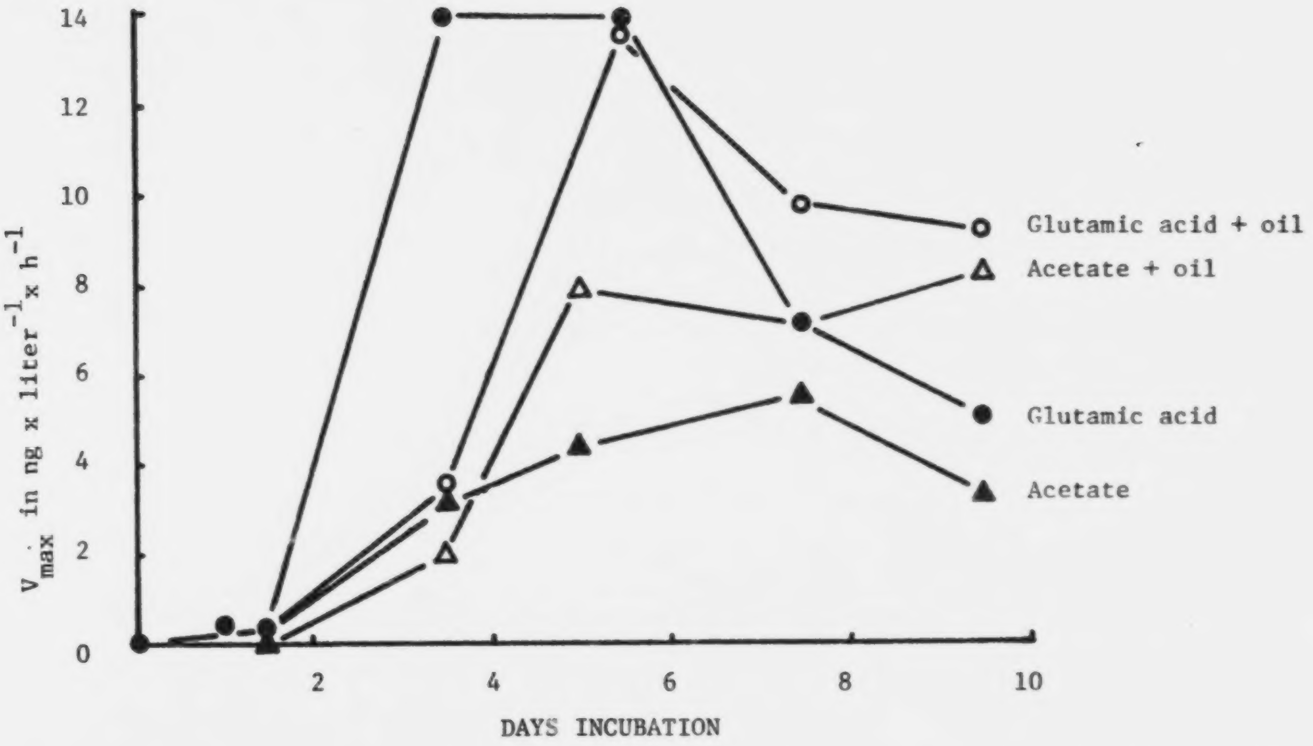

Figure 39. Effects of fresh Prudhoe Bay crude oil on relative microbial activity as measured with glutamic acid or acetate uptake.

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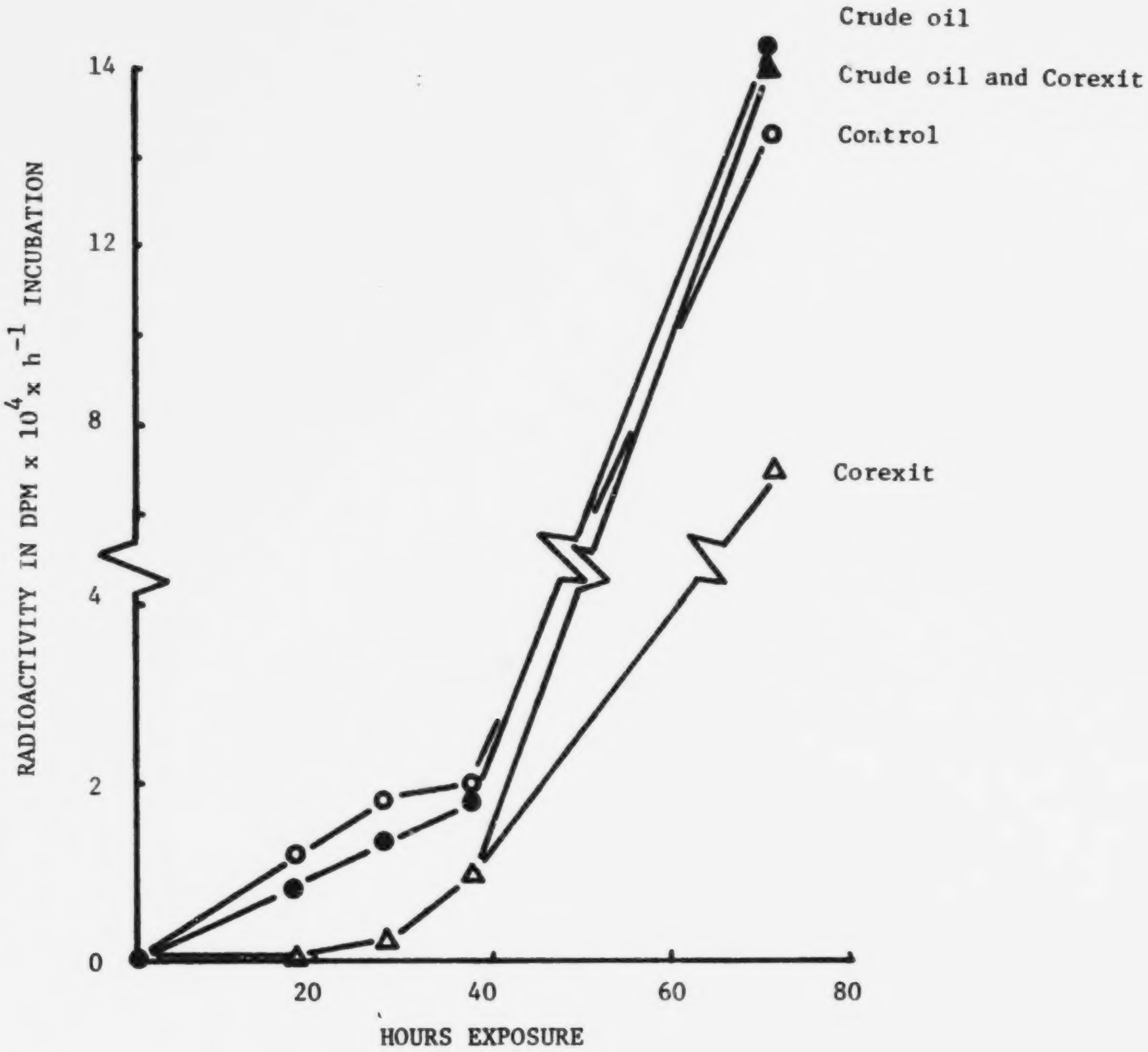

Figure 40. Effects of Cook Inlet crude oil on relative microbial activity measured by glucose uptake.

to the presence of crude oil but that the crude oil is providing a nutrient source for the organisms present.

Even though the glucose uptake rates were generally reduced in the presence of crude oil, a wide range of effects was observed and, in some cases, the uptake rate was actually higher in the treated sample (Table 17). The degree to which water samples were affected by the presence of crude oil was analyzed in terms of sample location. During the 1978 Cook Inlet cruise, a series of consecutive water samples was collected on a transect starting near Augustine Island and ending near Homer, Alaska (Fig. 41). The water samples collected in the center of the Inlet showed less effect than those collected at either end of the transect.

Water samples were also taken at two locations just north of Augustine Island. The one to the east was taken in Oil Bay which is so named because of a natural oil seep in that region. The one to the west was taken in a similar bay in which no seep has been reported. The reduction in glutamate uptake in the water sample taken from Oil Bay was only 12% as contrasted to a reduction of 67% observed at the other location (the reduction in the glucose acid uptake in the same samples was 0% and 61% respectively).

A series of observations was also made in water samples collected at three sample locations, one near Homer and two to the north near Kalgin Island. The range of values observed at these locations are illustrated in Figure 41. The percent reduction values observed in water samples collected at the northern stations are significantly lower than those observed in the station located near Homer.

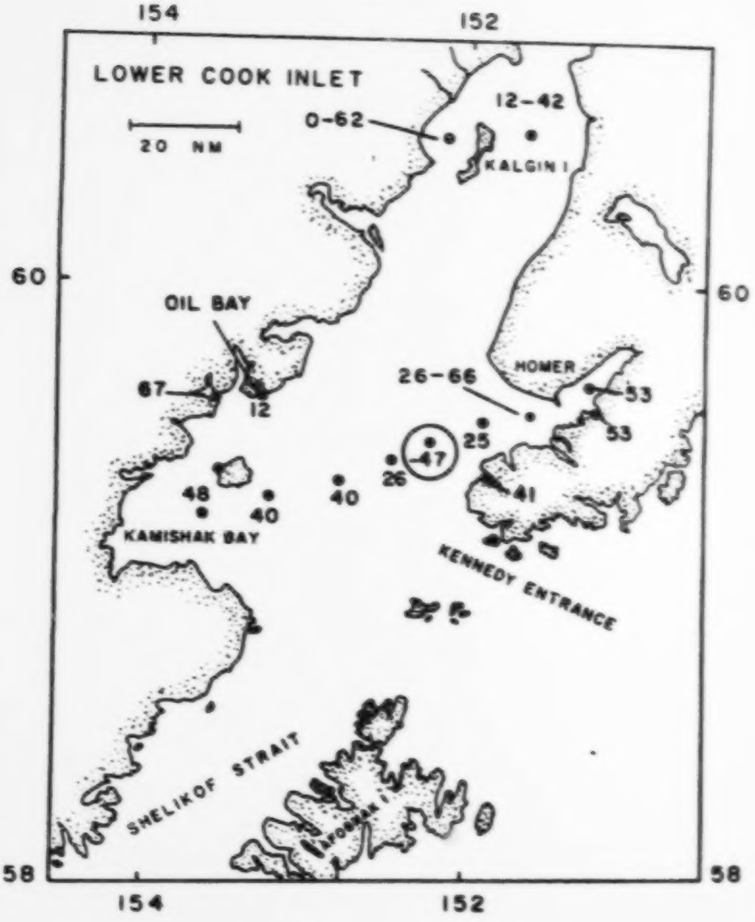
154
152
LOWER COOK INLET
20 NM
0-62
12-42
KALGIN I
60
60
OIL BAY
HOMER
26-66
67
12
53
53
25
-47
26
40
48
40
41
KAMISHAK BAY
KENNEDY ENTRANCE
SHELIKOF STRAIT
AFOGNAK I
58
58
154
152

Figure 41. Percent reduction in glutamate uptake in water samples exposed to crude oil during the April 1978 cruise. Circled station indicates location where sample showed increased glutamate uptake in the presence of crude oil.

b. Acute effects of crude oil on nitrogen fixation rates.

We conducted a series of experiments in which we added crude oil to sediments during the 24 hour incubation period used to measure nitrogen fixation. In some of the studies, we added sucrose to the reaction vessel to stimulate nitrogen fixation rates. The studies were conducted on samples collected from various locations within three regions (Table 19). A total of 75 sediment samples were analyzed during the course of this study. In none of the studies was there a statistical difference between the mean values observed.

2. Short-term effects of the dispersant Corexit 9527.

a. Effects on relative microbial activity and respiration percentages.

During 6 different sampling periods, a total of 149 water samples were studied to determine the effects of the crude oil dispersant Corexit 9527 and Corexit with crude oil on glucose uptake during the initial 8 hour exposure (Table 20). With the exception of the Beaufort Sea study, all water samples were exposed to 50 ppm Corexit. The mean value for the percent reduction in glucose uptake rates ranged from 58 to 95% in samples exposed to Corexit alone. When the microbial populations were exposed to Corexit and crude oil, the mean reduction ranged from 75 to 95%. In 3 of the 5 studies where the effects of both treatments were observed, the mean percent reduction in glucose uptake rates was higher in the samples treated with both Corexit and crude oil. These differences were very significant ($p<0.001$) in the Upper Cook Inlet and the July 1979 Kasitsna Bay studies.

Table 19. Acute effects of crude oil on nitrogen fixation rates.[1]

		Field Samples		Sucrose Added	
	n	Control	Oil	Control	Oil
Yaquina Bay	5	0.7	0.4	8.7	7.1
Cook Inlet, Apr. 1977	5	0.3	0.3	3.7	3.2
Cook Inlet, Nov. 1977	12	0.3	0.3	0.5	0.6
Cook Inlet, Apr. 1978	17	0.6	0.5	0.7	0.5
Beaufort Sea, Sept. 1977	9	0.1	0.1	0.5	0.4
Beaufort Sea, Jan. 1978	9	0.1	0.1	0.3	0.2
Beaufort Sea, Apr. 1978	5	1.3	1.2	-	-
Beaufort Sea, Aug. 1978	13	0.15	0.16	-	-

[1] mean rates for n samples expressed as ng N-fixed/g dry wt/hr.

Table 20. Percent reduction in glucose uptake rates in water samples.

	Treatment							
Sampling Location	Corexit				Corexit 9527 + Crude Oil			
	$\bar{y}$	SD	n	range	$\bar{y}$	SD	n	range
*Beaufort Sea	58	22	14	38-79	76	20	14	54-91
Cook Inlet	88	9	37	59-98	94	5	37	78-100
Norton Sound	85	11	60	58-98	-	-	-	-
Kasitsna Bay, 2/79	68	11	7	53-83	59	20	7	29-83
Kasitsna Bay, 4/79	95	3	6	92-98	95	5	6	85-99
Kasitsna Bay, 7/79	82	19	25	38-98	91	10	24	50-99

*These samples treated with 15 ppm Corexit, all other samples treated with 50 ppm Corexit.

The effect of Corexit and Corexit with crude oil on glucose uptake was also measured in sediment samples collected during the same sampling periods (Table 21). The mean percent reduction in the glucose uptake rates ranged from 15 to 60% in the presence of Corexit and from 40 to 79% in the presence of Corexit and crude oil. In four out of the five studies, the latter treatment showed a greater effect. These differences were statistically significant ($p<0.02$) in all cases. The effects of Corexit and Corexit with crude oil on glucose uptake rates were greater in water samples than in the sediments. For Corexit alone, the statistical significance of this difference was at the $p<.001$ level and it was at the $p<0.04$ level for the Corexit with crude oil treatments.

The above mentioned analyses were conducted using the same concentration of labeled glucose. It is possible that some component of the Corexit produced the same effect as adding non-labeled glucose to the reaction mixtures, thus reducing apparent uptake rates. One method that could be used to determine if this is taking place is to observe the effect of Corexit on the kinetics of glucose uptake using several concentrations and using the equations of Wright and Hobbie (1966). If Corexit is not acting as a metabolic inhibitor, the turnover time (T_t) and the transport constant plus the natural substrate concentration (K_t+S_n) values may change but the maximum potential velocity (V_{max}) of glucose uptake should not change. When this experiment was conducted in three water samples, the V_{max} values decreased in the presence of Corexit suggesting that it is acting as a metabolic inhibitor (Table 22). It should be noted that the incubation times of from 1

Table 21. Percent reduction in glucose uptake rates in sediment samples.

	Treatment							
Sampling Location	Corexit 9527				Corexit 9527 + Crude Oil			
	$\bar{y}$	SD	n	range	$\bar{y}$	SD	n	range
*Beaufort Sea	15	11	12	0-38	40	17	12	0-62
Cook Inlet	57	17	6	39-81	79	13	6	54-93
Norton Sound	54	13	34	30-84	-	-	-	-
Kasitsna Bay, 2/79	38	14	12	20-61	52	14	12	38-74
Kasitsna Bay, 4/79	44	24	11	17-75	44	24	11	6-76
Kasitsna Bay, 7/79	60	9	20	45-80	74	6	20	65-85

*These samples treated with 15 ppm Corexit, all other samples treated with 50 ppm Corexit.

Table 22. Effects of Corexit on the kinetics of glucose uptake in water samples.

	Control				Corexit			
Sample Number	Percent Respiration	V_{max}	K_t	T_t	Percent Respiration	V_{max}	K_t+S_n	T_t
313	22	0.30	5	19	16	0.90	8	84
314	29	0.016	0.2	13	15	0.0015	0.03	19
315	32	0.11	8	77	22	0.011	3	257

V_{max} = the maximum potential rate of glucose uptake reported as μg x $liter^{-1}$ x h^{-1}.

K_t+S_n = the transport constant plus the natural glucose concentration reported as μg/liter.

T_t = the time in hours required for the microbial population to utilize the naturally occurring glucose.

to 4 hours were used in these experiments. This indicates that Corexit probably affects glucose transport soon after exposure.

The effect of Corexit on percent respiration in both water and sediment samples was also observed (Tables 23 and 24). If the transport of glucose into the cells was the only function effected by Corexit, one would expect to see no changes in the percent respiration. There was a significant difference between the respiration percentages observed in treated and nontreated samples. For both water and sediment samples, 4 out of 6 studies showed significant differences in respiration percentages. Of those studies showing a significant difference in the sediment samples, all mean values increased in samples exposed to Corexit (Table 23). In the water samples, two showed increases (Table 24). In the Norton Sound study, there was a slight increase in the mean percent respiration in the treated samples but the difference was not statistically significant. If, however, the water samples were analyzed as two groups, one group showed an increase in the mean and the other showed the reverse trend (Fig. 42). The differences in both groups are very significant ($p<0.006$). The two sets of samples taken from two different locations in and near the Norton Sound are shown in Figure 31. The water samples in group A were collected to the west. Surface salinity and relative microbial activity measurements taken during the same cruise suggest that there are two water masses present in the area which are roughly defined by the line in Figure 31. The one to the west is more saline and shows lower levels of microbial activity than does the one to the east. These data suggest that different microbial populations may be affected in different ways by the presence of Corexit.

Table 23. Percent respiration observed in sediment samples treated with corexit.

	Control				Corexit 9527				
Sampling Location	$\bar{y}$	SD	n	range	$\bar{y}$	SD	n	range	*p =
Beaufort Sea	31	9	18	22-57	34	9	18	23-60	0.022
Cook Inlet	44	26	11	14-78	32	13	11	21-60	NS
Norton Sound	28	8	38	15-66	35	6	38	22-53	0.00003
Kasitsna Bay, 2/79	20	5	15	13-30	24	6	15	9-33	NS
Kasitsna Bay, 4/79	17	8	12	6-34	18	5	12	9-25	0.007
Kasitsna Bay, 7/79	18	3	20	12-23	23	5	20	16-34	0.000008

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*Level of statistical significance between treated and non-treated samples.

Table 24. Percent respiration observed in water samples treated with Corexit.

	Control				Corexit 9527				
Sampling Location	$\bar{Y}$	SD	n	range	$\bar{Y}$	SD	n	range	*p =
Beaufort Sea	40	14	13	18-58	28	11	13	15-51	0.0005
Cook Inlet	28	15	39	11-70	32	13	39	14-67	0.013
Norton Sound	26	7	62	15-49	28	7	62	11-44	NS
Kasitsna Bay, 2/79	32	3	6	28-38	35	7	6	29-46	0.040
Kasitsna Bay, 4/79	37	7	7	28-46	47	10	7	34-62	NS
Kasitsna Bay, 7/79	34	9	24	21-50	26	10	24	13-53	0.007
Norton Sound Group A	24	6	48	11-49	28	5	48	18-42	0.00001
Norton Sound Group B	34	8	14	20-46	25	10	14	11-44	0.0006

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*Level of statistical significance between treated and non-treated samples.

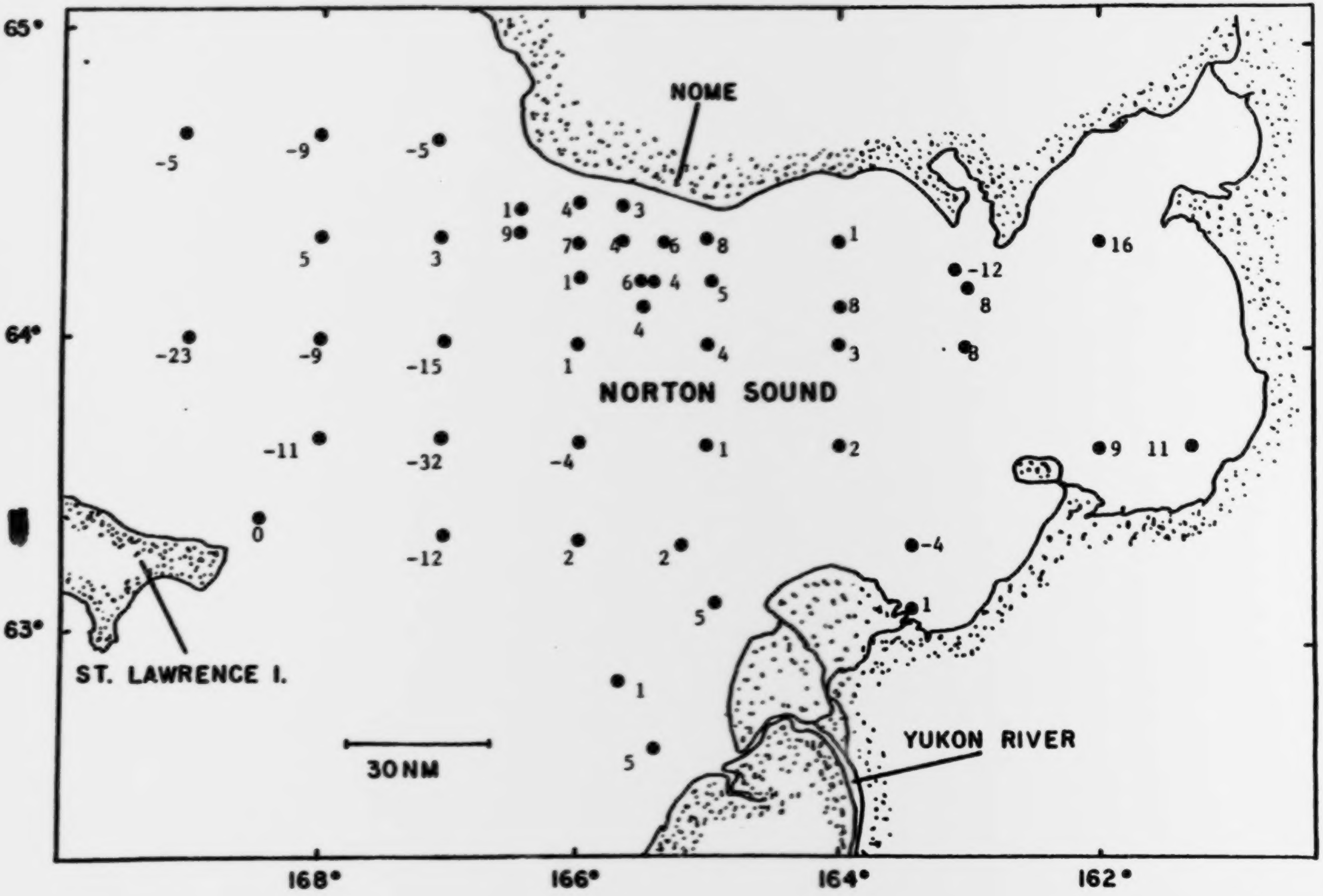
65°
64°
63°
168°
166°
164°
162°
NOME
NORTON SOUND
ST. LAWRENCE I.
YUKON RIVER
30NM
-5
-9
-5
1
4
3
9
5
3
7
4
6
8
1
16
-12
1
6
4
5
8
4
8
-23
-9
-15
1
4
3
8
-11
-32
-4
1
2
9
11
0
-12
2
2
-4
5
2
1
1
5

Figure 42. Change in percent respiration of glucose in water samples exposed to 50 ppm Corexit.

The relationship between Corexit concentration and its effect on glucose uptake in 9 Kasitsna Bay water samples was studied using three concentration ranges: 0-100, 0-50, and 0-20 ppm (Table 25). Figure 43 graphically illustrates the results of one of the three studies. Both this study and the one conducted over a range of from 0-100 ppm indicate that most of the effect takes place at concentrations below 20 ppm with little further change observed at 50 and 100 ppm. In the 0-20 ppm study, the samples showed decreases in uptake rates of 10 and 12% when exposed to Corexit at 1 ppm. Using the best fitting power curve to describe the data, the mean concentration at which 50% of the original uptake rate was lost was calculated to be 12 ppm. These data suggest that the alterations in microbial activity that were observed in the 50 ppm studies would represent the maximum effect possible.

b. The effects of Corexit 9527 on glucose uptake and respiration with exposure times up to 5 days.

In three studies, we observed the effects of Corexit on glucose uptake and respiration for periods of time greater than 8 hours and less than 130 hours. In the shortest study (Fig. 40), the effects of Corexit and Corexit with crude oil were observed for periods of time up to 72 h. During the entire study, the glucose uptake rates by pelagic microorganisms were lower in the Corexit-treated sample than in the control. The same was true when the water samples were exposed to Corexit and crude oil for up to 38 h; however, after 72 h, the rate of uptake in this water sample was actually greater than that observed in the control. In the other two studies, longer exposure times were used. In one experiment (Fig. 44), the uptake rates observed

Table 25. The concentration of Corexit at which the glucose uptake rate was reduced to one half of the control.

Sample Number	Corexit Concentration ppm	Goodness of Fit to Power Curve	Range of Concentrations Tested - ppm
271	15	0.94	0-100
273	18	0.96	0-100
275	11	0.92	0-100
285A	7	0.99	0-20
285B	7	0.99	0-20
242	10	0.98	0-50
247	10	0.82	0-50
250	10	0.98	0-50
255	10	0.98	0-50

mean = 12 ppm (to reduce uptake 50%)

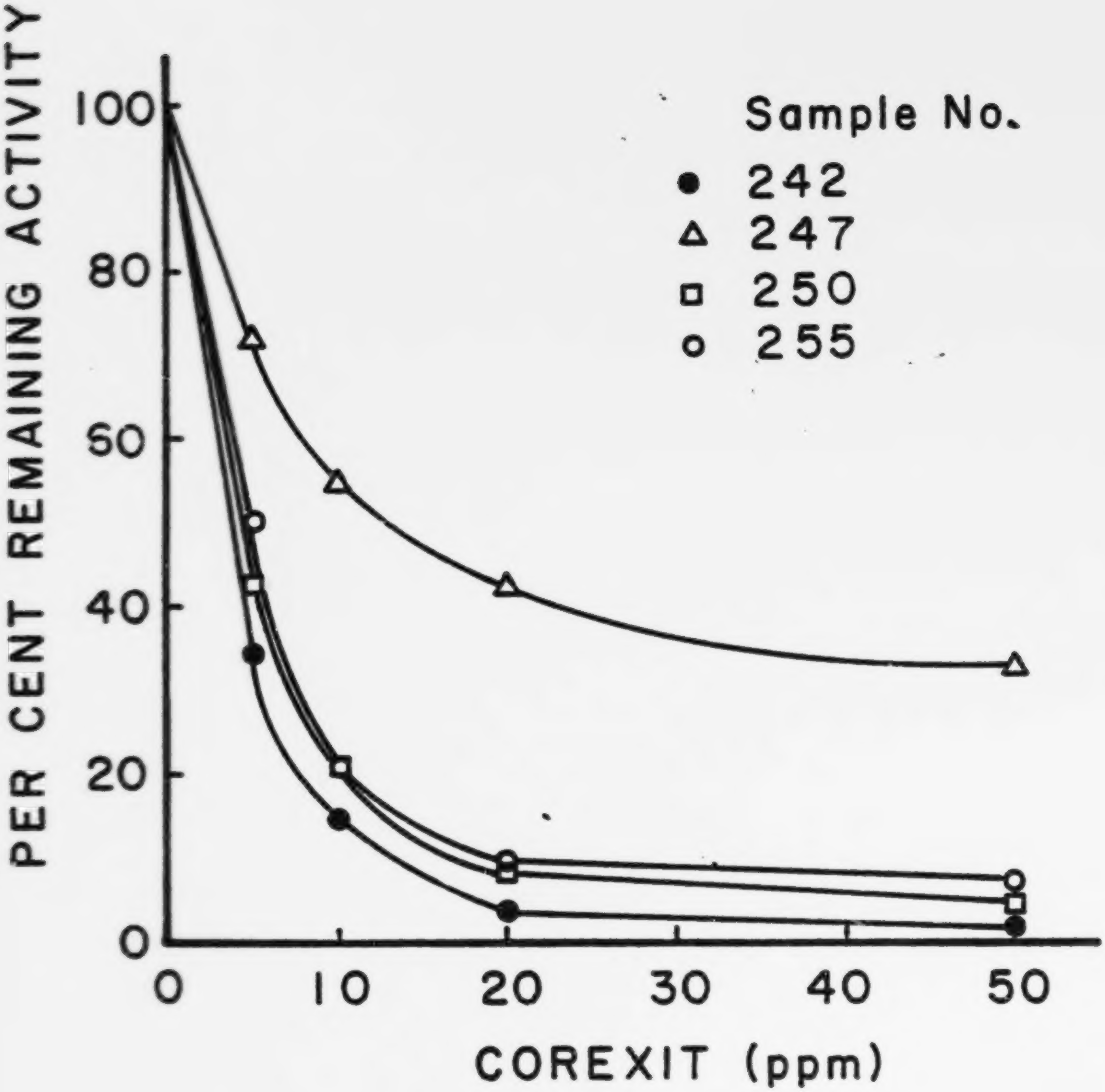

Figure 43. Changes in glucose uptake relative to controls in four water samples exposed to increasing concentrations of Corexit.

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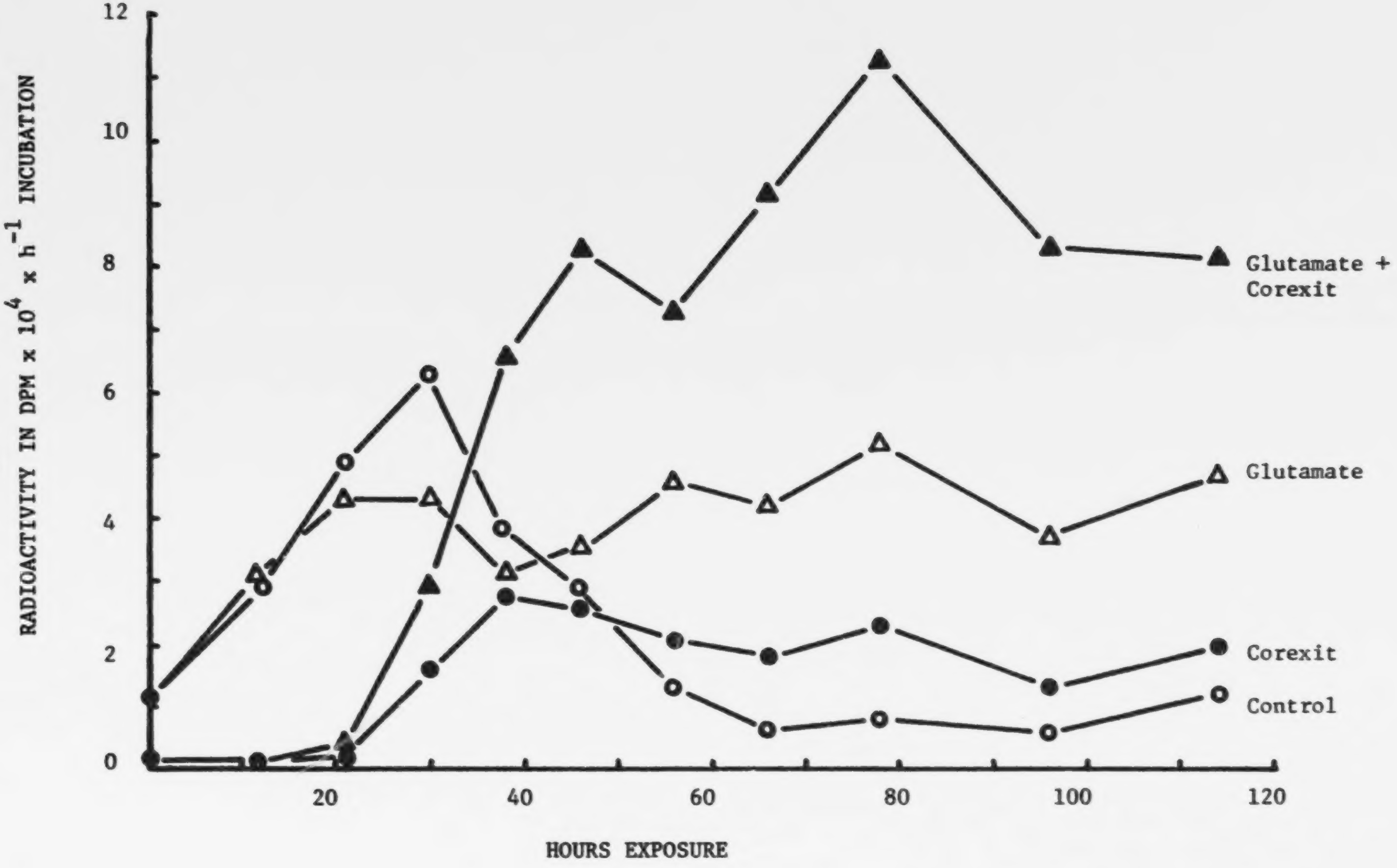

Figure 44. The effects of Corexit 9527 and glutamic acid on glucose uptake by pelagic microorganisms.

in the Corexit-treated sample were greater than the control. During the same experiment, glucose uptake rates were also monitored in water samples amended with either non-labeled glutamic acid or glutamic acid and Corexit. Samples treated in these ways showed increased activity relative to the control after about 45 h incubation. The result of this experiment suggested to us that the metabolism of Corexit may be limited by the presence of fixed nitrogen. When we repeated the experiment, we also processed replicate water samples to which either NH_4^+ or NH_4^+ and Corexit had been added (Fig. 45). The results were essentially the same as those observed in the earlier experiment except the time for the "recovery" in the Corexit-treated samples was shorter than before. When comparing the Corexit and Corexit with NH_4^+ treatments, it was found that after about 40 h exposure, the water sample to which Corexit and NH_4^+ was added showed higher uptake rates than the sample treated with Corexit alone.

During the same experiment, we challenged (exposed) subsamples with Corexit to determine what effect Corexit might have on glucose uptake rates in samples treated in various ways. In the first experiment, this was done after an incubation period of 114 h (Fig. 44). Of the three treatments, none of these showed a significant decline in glucose uptake rates when challenged to Corexit; however, a reduction of 65% was seen in the control. The results were the same in the second experiment where the only significant reduction was observed in the control (75%) and the NH_4^+ treated water (85%) after 92 h exposure.

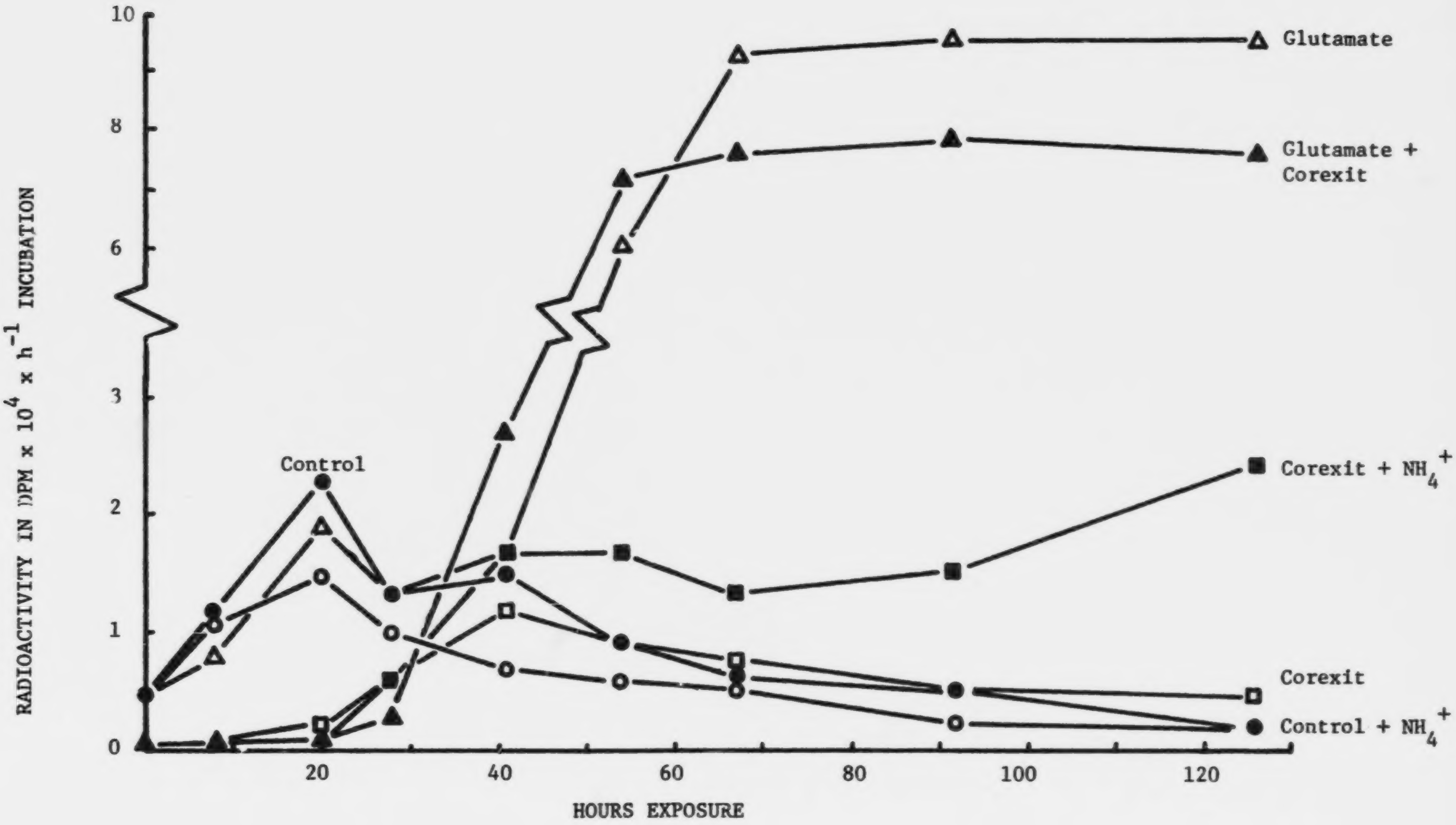

Figure 45. The effects of Corexit, NH_4^+, and glutamic acid on glucose uptake by pelagic microorganisms.

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595

An analysis of the percent respiration during the same experiments showed that by the end of the experiment, the percent respiration was higher in the Corexit-treated samples than in the controls (Fig. 46). The lowest respiration percentages were observed in those samples to which glutamic acid was added (with or without Corexit).

c. Corexit and crude oil effects on bacteriovorous activity by a marine ciliate.

The ingestion of bacterial cells by protozoa in the presence of crude oil and/or Corexit 9527, is expressed as a percentage of the control treatment (Table 26). Crude oil, at a concentration sufficient to produce a surface slick, caused slight but significant ($p<0.01$) decrease. This concurs with the observations of Andrews and Floodgate (1974) that protozoa are essentially unharmed by the presence of crude oil and may even take up droplets concurrent with feeding activities. When 15 ppm Corexit 9527 was added to the crude oil the ingestion of bacteria was decreased approximately 20-fold. Corexit (15 ppm) alone produced almost a 10-fold reduction in ingestion. Venezia and Fossato (1977) point out that many of the initial reports of toxicity of crude oil and dispersants are probably due to increased contact with the oil rather than any significant effect of the dispersant itself. Our results indicate that in addition to accentuating the effects of crude oil, Corexit 9527 alone has a detrimental effect on the feeding of the marine ciliate tested. The combined action of crude oil and corexit appears to be synergistic since the observed ingestion of bacterial cells is about half the value expected if the effects of crude oil and Corexit were independent (4.9 vs 10.1%).

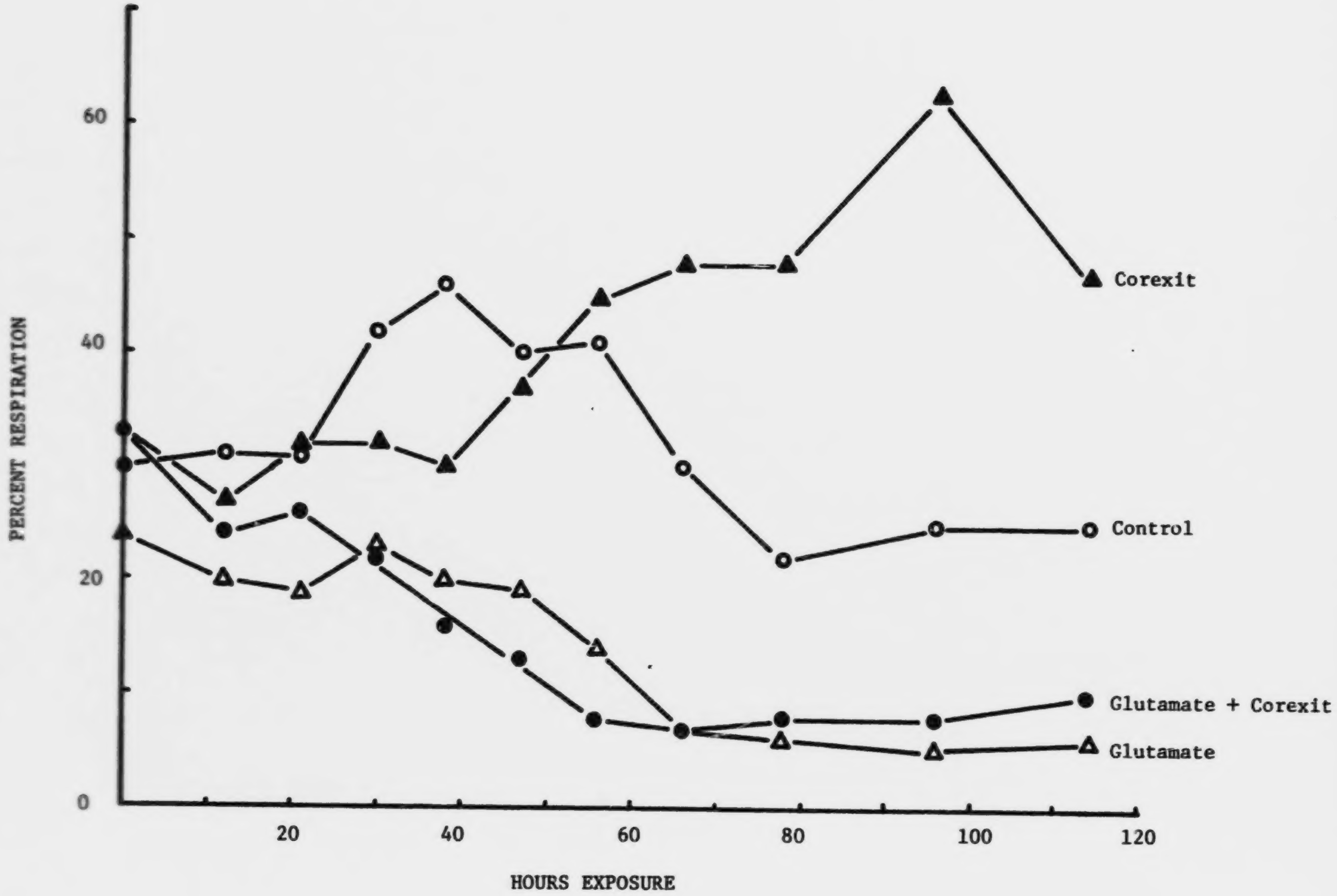

Figure 46. The effects of Corexit 9527 and glutamic acid on percent respiration by pelagic microorganisms.

597 BEST DOCUMENT AVAILABLE

Table 26. Ingestion of *Vibrio alginolyticus* cells by a marine ciliate when exposed to crude oil and/or Corexit.

Treatment	PERCENT INJESTION CELLS[a]	p-VALUE[b]
Control[c]	100.0 ± 2.9	
Oil (10 ppm)	82.7 ± 5.2	0.01
Oil + Corexit (15 ppm)	4.9 ± 1.0	0.001
Corexit 9527 1 ppm[d]	75.3 ± 5.4	0.01
5 ppm[d]	51.1 ± 2.5	0.001
10 ppm[d]	27.2 ± 3.1	0.001
15 ppm[d]	12.2 ± 2.5	0.001
50 ppm[d]	2.3 ± 0.9	0.001
100 ppm[d]	1.7 ± 0.3	0.001

---

[a] percent ingestion compared to control with mean ± standard deviation.

[b] significance level (p) of treatment compard with control.

[c] control arbitrarily set at 100%.

[d] power curve equation for Corexit concentrations 1 to 100 ppm:

$$Y \text{ (percent ingestion)} - 134 \times (\text{ppm})^{-0.923}$$

When concentrations of Corexit 9527 were increased from 1 to 100 ppm, there were significant decreases in the uptake of bacterial cells. For Corexit treatments in the 1 to 50 ppm range the effects of each concentration was also significantly decreased ($p<0.01$) from the preceeding concentration (i.e., % uptake control>1 ppm>5>10>15>50). The depression in ingestion rates observed at 50 ppm was essentially the same as that observed at 100 ppm.

It should be noted that the levels of Corexit 9527 used in this study were high relative to levels anticipated under actual field application in open waters (Lindblom, 1978). In one study where concentrations of a related Corexit compound were monitored under these conditions, the highest reported concentration was 1 ppm (McAuliffe et al., 1975). This does not however, rule out the possibility that higher concentrations may be encountered when this dispersant is applied in the near-shore environment; especially in areas where there is little vertical mixing or current activity. Under these conditions it is conceivable that much higher concentrations may be found which would result in reduced cropping rates of bacteria by protozoa. This, in turn, would result in reduced rates of carbon and energy transfer through the detrital food chain.

B. Long-term effects of crude oil and/or Corexit on microbial activities in marine sediments.

We have participated in two long-term effects studies, one which was initiated in July 1977 in Elson Lagoon (near Point Barrow, AK) and the other which was initiated in February 1979 at Kasitsna Bay (near Homer, AK in Cook Inlet).

1. Effects on relative microbial activity as measured by glucose, glutamic acid and acetate uptake.

a. The Kasitsna Bay study

During the Kasitsna Bay study, we participated in 6 field study periods after the study was initiated in Feb. 1979: April 1979 and 1980; July-August 1979 and 1980; October-November 1979; and January 1980. In February 1979, we initiated the first long-term effects study where treated and nontreated sediments were placed in trays which were subsequently placed on the bottom of Kasitsna Bay by SCUBA divers. Portions of these sediments were also placed in aquaria fitted with a seawater flow system. A large number of variables were monitored over the 1.5 year lifespan of the study including the effects of crude oil on relative microbial activity (Tables 27-31). Observations were made after 1.5, 5, 8, 11, and 18 months exposure. Throughout the study, relative microbial activity was monitored by following the uptake and respiration of labeled glucose and glutamic acid. In the last two field periods (11 and 18 mo exposure) relative microbial activity was also monitored using labeled acetate. With the exception of the 8 month exposure observations, the presence of fresh Cook Inlet crude oil at 50 ppt significantly reduced relative microbial activity as measured using the above mentioned substrates. The percent reduction observed in treated sediments at the first observation period (1.5 mo) was 50 and 73 respectively for glucose and glutamic acid. At the end of the study (18 mo) these figures were reduced to 42 and 40%.

During the course of our Kasitsna Bay work, we also initiated studies which were designed to determine the effects of various concentrations of fresh crude oil, the time required for the observed changes to take place, the effects weathered crude oil on microbial

Table 27. Changes observed in sediment samples exposed to crude oil for 6 weeks.

	Trays				Aquaria				
	Control	Oil	Dif.	%	Control	Oil	Dif.	%	$p \leq$
Variable	$\bar{Y}$	$\bar{Y}$	Δ	Δ	$\bar{Y}$	$\bar{Y}$	Δ	Δ	
Glucose uptake	22	11	11	-50	20	13	7	-35	0.005
Glutamate uptake	335	92	243	-73	257	116	141	-55	0.0001
Glutamate Vmax	430	160	270	-63	300	176	124	-41	0.002
Glutamate Kt+Sn	20	14	6	-30	11	28	17	+155	NS
Glutamate Tt	85	135	50	+59	54	227	173	+320	0.03
Glucose % Resp.	20	38	18	+90	23	45	22	+110	0.0001
Glutamate % Resp.	44	64	20	+45	48	65	17	+26	0.00001
Nitrogen Fixation	0.6	0	0.6	-100	2.3	0.2	2.1	-91	0.004
Total Adenylates	4.5	2.2	2.3	-51	3.8	1.4	2.4	-63	0.0001
Energy Charge	0.33	0.26	0.07	-21	0.34	0.24	0.10	-29	
Hydrogen Ion Conc.	1.4	3.5	2.1	+150	0.5	3.1	2.6	+520	0.01
Redox Potential (Surface)	90	20	70	-78	18	-229	247	-1372	0.03

Under each heading, the mean value measured in the control (non-treated) and the oiled sediments is reported along with the difference between the means and the percent of that difference relative to the control mean. The "p" value is the level of statistical significance between the mean values of all control and treated sediments analyzed.

	Units		Units
Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}	Glucose % Resp.	%
Glutamate uptake	" " "	Glutamate % Resp.	%
Glutamate Vmax	" " "	Nitrogen Fixation	ngN$_2$ x g. dry wt.$^{-1}$ x h^{-1}
Glutamate Kt+Sn	μg x liter^{-1}	Total Adenylates	μM x g. dry wt.$^{-1}$
Glutamate Tt	h.	Energy charge	ratio - see methods
		Hydrogen ion conc.	x E-8 M
		Redox potential	mV

Table 28. Changes observed in sediment samples exposed to crude oil for 5 months.

	Trays				Aquaria				
	Control	Oil	Dif.	%	Control	Oil	Dif.	%	p≤
Variable	$\bar{Y}$	$\bar{Y}$	Δ	Δ	$\bar{Y}$	$\bar{Y}$	Δ	Δ	
Glucose uptake	77	31	46	-60	140	44	96	-69	0.0001
Glutamate uptake	605	178	427	-71	815	221	594	-73	0.0001
Glutamate Vmax	1040	232	808	-78	1060	300	760	-72	0.001
Glutamate Kt+Sn	44	18	26	-60	18	17	1	-6	0.01
Glutamate Tt	60	130	70	+117	26	84	58	+223	0.003
Glucose % Resp.	14	51	37	+264	21	49	28	+133	0.00001
Glutamate % Resp.	46	50	4	+9	49	42	7	+14	0.007
Nitrogen Fixation	3.4	0.5	2.9	-85	2.0	0.1	1.9	-95	0.00002
Total Adenylates	5.6	1.5	4.1	-74	1.7	0.6	1.1	-65	0.005
Energy Charge	0.34	0.23	0.09	-26	0.39	0.32	0.07	-18	NS
Hydrogen Ion Conc.	1.5	4.3	2.8	+187	2.9	4.2	1.3	+45	0.0005
Redox Potential	85	-238	323	-380	61	-290	351	-576	0.000001

Under each heading, the mean value measured in the control (non-treated and the oiled sediments is reported along with the difference between the means and the percent of that difference relative to the control mean. The "p" value is the level of statistical significance between the mean values of all control and treated sediments analyzed.

	Units
Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}
Glutamate uptake	" " "
Glutamate Vmax	" " "
Glutamate Kt+Sn	μg x liter^{-1}
Glutamate Tt	h.

	Units
Glucose % Resp.	%
Glutamate % Resp.	%
Nitrogen Fixation	ng x g. dry wt.$^{-1}$ x h^{-1}
Total Adenylates	uM x g. dry wt.$^{-1}$
Energy charge	ratio - see methods
Hydrogen ion conc.	x E-8 M
Redox potential	mV

Table 29. Changes observed in sediment samples exposed to crude oil for 8 months.

	Trays and Aquaria				
	Control	Oil	Dif.	%	p≤
Variable	$\bar{Y}$	$\bar{Y}$	Δ	Δ	
Glucose uptake	55	34	-11	-38	NS (0.27)
Glutamate uptake	448	353	-95	-21	NS
Glucose % Resp.	16	35	19	+119	0.003
Glutamate % Resp.	46	49	3	+7	NS
Nitrogen Fixation	3.5	0.2	-3.3	-95	0.003
Denitrification	92	45	-47	-51	0.009
CO$_2$ Production	1.3	2.6	1.3	+50	0.002
Hydrogen Ion Conc.	2.7	5.4	2.7	+50	NS
Redox Potential	+33	-71	104	-315	NS (0.07)

Under each heading, the mean value measured in the control (non-treated) and the oiled sediments is reported along with the difference between the means and the percent of that difference relative to the control mean. The "p" value is the level of statistical significance between the mean values of all control and treated sediments analyzed.

	Units		Units
Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}	Hydrogen ion conc.	x E-8
Glutamate uptake	" " "	Redox potential	mV
Nitrogen Fixation	ng x g. dry wt.$^{-1}$ x h^{-1}		
Denitrification	" " "		
CO$_2$ production	nM " "		

Table 30. Changes observed in sediment samples exposed to crude oil for 11 months.

	Trays and Aquaria				
	Control	Oil	Dif.	%	$p \leq$
Variable	$\bar{Y}$	$\bar{Y}$	Δ	Δ	
Acetate Uptake	54	14	40	-74	0.02
Glucose Uptake	36	18	18	-50	0.05
Glutamate Uptake	246	90	156	-63	0.002
Glutamate Vmax	237	89	148	-62	0.02
Glutamate Kt+Sn	33	24	9	-27	NS
Glutamate Tt	74	205	131	+177	0.01
Acetate % Resp.	23	38	15	+65	0.005
Glucose % Resp.	19	34	15	+79	0.0003
Glutamate % Resp.	49	55	6	+12	NS (0.07)
N_2 Fixation	0.56	.04	.52	-93	0.0003
Denitrification					
Natural	0.08	0	0.08	-100	0.004
Potential	95	25	70	-74	0.0004
CO_2 Production	5	12	7	+58	0.015
Methane Production	23	54	31	+57	0.04

Under each heading, the mean value measured in the control (non-treated) and the oiled sediments is reported along with the difference between the means and the percent of that difference relative to the control mean. The "p" value is the level of statistical significance between the mean values of all control and treated sediments analyzed.

Units

Acetate uptake	ng x g. dry wt.$^{-1}$ x h^{-1}	Nitrogen Fixation	ng x g dry wt.$^{-1}$ x h^{-1}
Glucose uptake	" " " " "	Denitrification	ng x g dry wt.$^{-1}$ x h^{-1}
Glutamate uptake	" " " " "	CO_2 production	nM " "
Glutamate Vmax	" " " " "	Methane production	pM " "
Glutamate Kt+Sn	µg x liter^{-1}		
Glutamate Tt	h.		

Table 31. Changes observed in sediment samples exposed to crude oil for 18 months.

	Trays only				
	Control	Oil	Dif.	%	P≤
Variable	$\bar{Y}$	$\bar{Y}$			
Acetate Uptake	38	24	14	-37	0.05
Glucose Uptake	113	65	48	-42	0.05
Glutamate Uptake	1223	728	495	-40	0.05
Glutamate Vmax	1390	440	950	-66	0.05
Glutamate Kt+Sn	20	13	7	-35	NS
Glutamate Tt	23	27	4	+17	NS
Acetate % Resp.	31	37	6	+10	NS
Glucose % Resp.	22	40	18	+45	0.001
Glutamate % Resp.	47	54	7	+17	0.05
Ethylene Production	2.9	0.7	2.2	-76	0.001
Denitrification	170	45	125	-73	0.01
CO_2 Production	15	26	11	+73	NS
Phosphatase Activity	0.38	0.24	0.14	-37	0.001
Arylsulfatase Activity	0.70	0.42	0.29	-41	0.001
Methane Production	2.2	6.3	4.1	+186	0.01
Hydrogen Ion Concentration	3.4	6.2	2.8	+82	0.05
Redox Potential (Bottom)	-207	-447	240	-116	0.001
Direct Counts	5.82	5.13	.69	-12	NS

	Units		Units
Acetate uptake	ng x g. dry wt.$^{-1}$ x h^{-1}	Denitrification	ng x g. dry wt.$^{-1}$ x h^{-1}
Glucose uptake	" " " " "	CO_2 Production	nM x g. dry wt.$^{-1}$ x h^{-1}
Glutamate uptake	" " " " "	Phosphatase activity	uM PNP x g. dry wt.$^{-1}$ x h^{-1}
Glutamate Vmax	" " " " "	Arylsulfatase activity	uM PNP x g. dry wt.$^{-1}$ x h^{-1}
Glutamate Kt+Sn	ug x liter^{-1}	Methane production	pM x g. dry wt.$^{-1}$ x h^{-1}
Glutamate Tt	h.	Hydrogen Ion conc.	x 10^{-8}
% Respiration	%	Redox potential	mV
Nitrogen fixation	ng x g. dry wt.$^{-1}$ x h^{-1}	Direct counts	x 10^{8} x g. dry wt.$^{-1}$

function, and the effects of overlaying nontreated sediment with sediment treated with both fresh and weathered crude oil. We conducted one study in which we observed the effects of fresh crude oil at various concentrations after exposure for 1, 3, and 12 months (Fig. 47). In all three of these studies, we found the relative microbial activity was decreased with increasing concentrations up to 1 ppt. When the concentrations were increased above that level, there was little or no additional reduction in activity.

In another study, we observed the effects of 50 ppt crude oil on relative microbial activity in three sediments exposed for various times (Fig. 48). We observed that most of the effect took place within the first 1.5 days exposure.

We also considered the effects of "weathered" crude oil on relative microbial activity (Table 32). In this study, we found that "weathered" crude oil concentrations of 50 ppt reduced activity by 63% ($p<0.001$) after a one year exposure and it reduced activity by 47% ($p<0.01$) at a concentration of 1 ppt.

In addition to this study, we also conducted a series of observations on the effects of both fresh and "weathered" crude oil overlays on nontreated sediments (Table 33). In this study, the oil concentration was 50 ppt and the exposure time was one year. The samples were placed either in aquaria (one control and one weathered crude treatment) or in trays (two trays each containing nontreated sediments, weathered and fresh crude for a total of six trays). In the aquaria experiment, the relative microbial activity was reduced by 21% in the treated sample but his reduction was not significant (replicate observations were made in samples taken from either side of the partitioned down the center of the aquaria). In the tray experiment, there was no significant

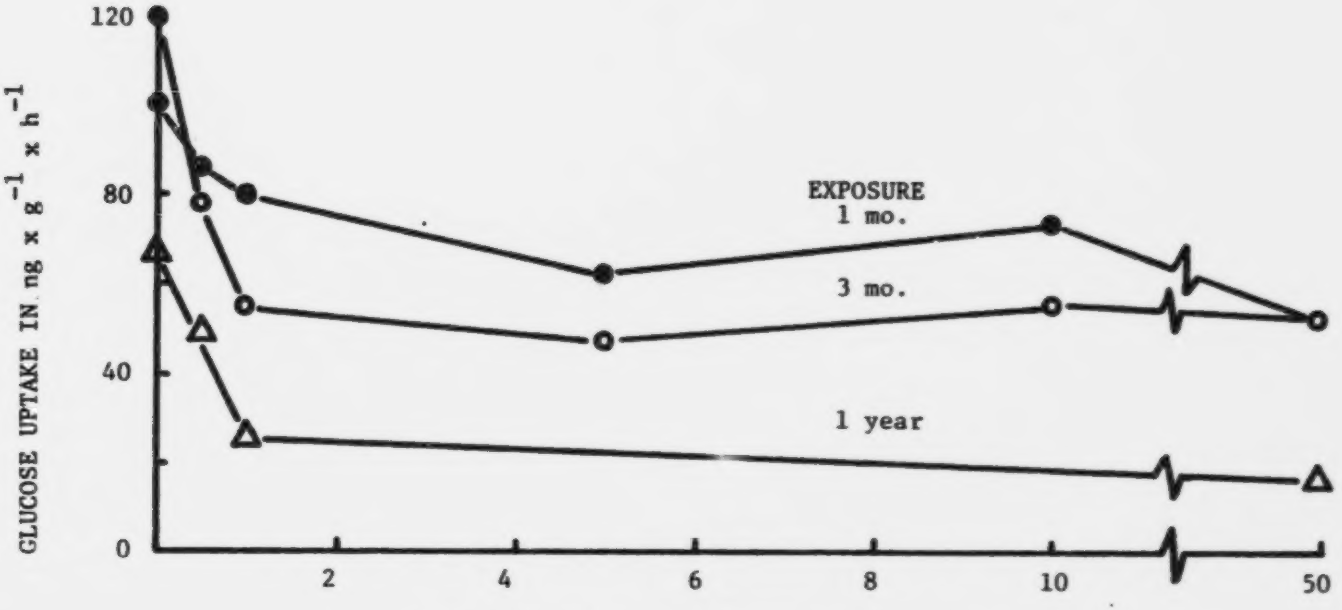

Figure 47. Effects of various concentrations of fresh crude oil on glucose uptake rates in three marine sediments.

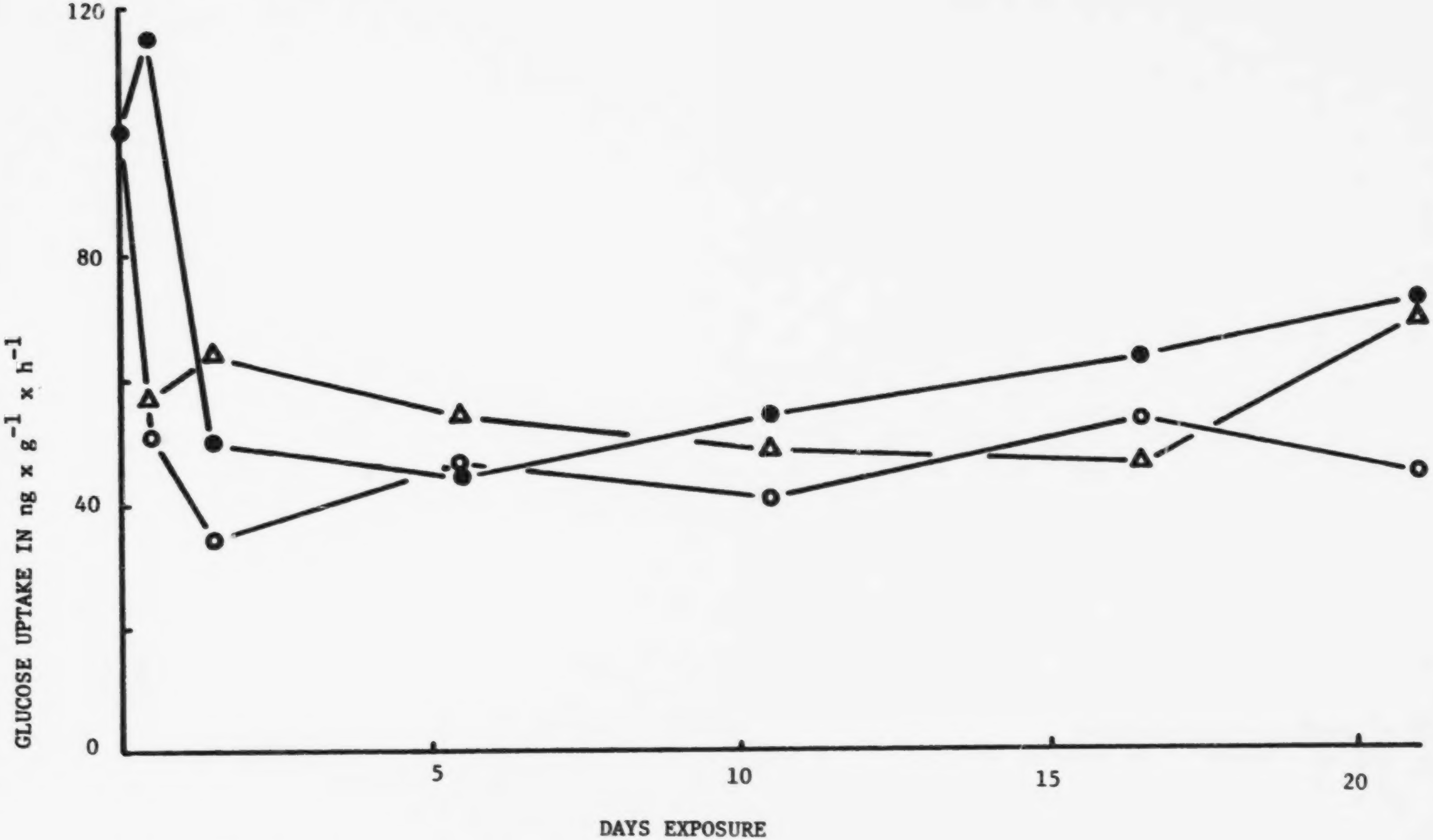

Figure 48. Effects of fresh crude oil at 50 ppt on glucose uptake rates in three marine sediments at various exposure times.

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Table 32. Effects of various concentrations of fresh and "weathered" crude oil on microbial activities in a marine sediment exposed for one year.

		Control	Fresh			Weathered	
			50 ppt	1.0 ppt	0.1 ppt	50 ppt	1.0 ppt
Glucose uptake	$\bar{Y}$	68	17	26	49	25	39
	%Δ		-75	-62	-28	-63	-47
	p		***	**	NS	***	**
% Respiration	$\bar{Y}$	21	38	27	24	32	20
	%Δ		+81	+29	+14	+52	-5
	p		**	*	NS	**	NS
Methane Production	$\bar{Y}$	1.8	4.5	3.2	2.4	3.4	2.3
	%Δ		+150	+78	+33	+89	+28
	p		*	NS	NS	**	NS
Nitrogen Fixation	$\bar{Y}$	1.7	0.3	0.8	1.5	1.8	1.6
	%Δ		-82	-53	-12	+6	-6
	p		**	**	NS	NS	NS
Denitrification	$\bar{Y}$	135	43	88	115	42	115
	%Δ		-68	-35	-15	-69	-15
	p		**	*	NS	*	NS
Phosphatase Activity	$\bar{Y}$	251	159	195	NOT	234	392
	%Δ		-37	-22	RUN	-7	+56
	p		*	NS		NS	**
Redox Potential	$\bar{Y}$	-119	-395	-323	-225	-402	-289
	%Δ		-232	-171	-89	-238	-143
	p		***	**	*	***	**

Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}
Methane production	pM x g. dry wt.$^{-1}$ x h^{-1}
Nitrogen fixation	ng x g. dry wt.$^{-1}$ x h^{-1}
Denitrification	" " " "
Phosphatase	µM PNP x g. dry wt.$^{-1}$ x h^{-1}
Redox	mV

Statistical significance
NS = not significant
* = $p \leq 0.05$
** = $p \leq 0.01$
*** = $p \leq 0.001$

Table 33. Effects of fresh and "weathered" crude oil overlays on microbial activities in a marine sediment in trays or aquaria exposed for one year.

		TRAYS			AQUARIA	
		Control	Fresh	Weathered	Control	Weathered
CO_2 Evolution	$\bar{Y}$	21.4	14.1	21.0	5.4	11.7
	% Diff		-34	-2		+117
	P		NS	NS		
Nitrogen Fixation	$\bar{Y}$	2.6	1.2	3.5	2.1	5.1
	% Diff		-54	+26		+143
	P		**	NS		
Methane Production	$\bar{Y}$	2.7	3.4	1.8	2.2	3.0
	% Diff		+29	-34		+38
	P		NS	NS		NS
Glucose Uptake	$\bar{Y}$	107	37	121	56	44
	% Diff		-65	+13		-21
	P		*	NS		NS
% Respiration	$\bar{Y}$	23	29	24	21	25
	% Diff		+27	+4		+19
	P		*	NS		NS
Denitrification	$\bar{Y}$	156	122	44	158	145
	% Diff		-22	-72		-8
	P		*	**		NS
Redox Potential (Bottom)	$\bar{Y}$	-284	-359	-413	-232	-394
	% Diff		-26	-45		-70
	P		NS	NS		**

CO_2 Evolution	nM x g. dry wt.$^{-1}$ x h^{-1}
Nitrogen Fixation	ng x g. dry wt.$^{-1}$ x h^{-1}
Methane Production	pM x g. dry wt.$^{-1}$ x h^{-1}
Glucose Uptake	ng x g. dry wt.$^{-1}$ x h^{-1}
% Respiration	%
Denitrification	ng x g. dry wt.$^{-1}$ x h^{-1}
Redox Potential	mV

Statistical Significance		
	NS	Not Significant
	*	$P < 0.05$
	**	$P < 0.01$
	***	$P < 0.001$

reduction in microbial activity in the weathered crude overlay but there was in the fresh crude oil overlay (reduced 65% with $p<0.05$).

We were also curious about what changes might be found in sediments taken from another area near Kasitsna Bay. In November 1979, we initiated a study of the effects of fresh crude oil (50 ppt) on microbial function in sediments collected near the north shore of Kachamak Bay close to Homer, AK (Table 34). After treatment, these sediments were left at the location from which they were collected for 8 months before they were analyzed. In this study we observed a highly significant 61% reduction in relative microbial activity (glucose uptake rate).

In addition to considering the effects of crude oil on relative microbial activity, we also measured the effects of Corexit 9527 (500 ppm) and Corexit and crude oil on this and other variables. After an exposure period of 5 months, we analyzed both trays and aquaria which contained treated sediments (Table 35). In general, the changes that were observed in the sediments treated with both oil and Corexit showed greater effects than those treated with oil alone; however, these differences were not statistically significant. The sediments that were treated with Corexit alone showed little or no change when compared with the controls. After 18 months incubation, essentially no differences were observed between sediments treated with Corexit compared to those that had not been treated.

The study in which we observed the effects of crude oil and/or Corexit on relative microbial activity in pelagic populations, indicated that there may be an adjustment of these populations to the presence of these pollutants. We also wanted to determine what changes occurred in benthic populations exposed to these same pollutants for an extended period of time. In order to do this,

Table 34. Changes observed in Coal Bay sediment samples exposed to fresh crude oil for 8 months.

Variable	Control	Oil	% Difference	p ≤
Glucose uptake	158	54	-61	0.001
Glucose % resp.	23	42	+85	0.001
Nitrogen fixation	0.95	0.03	-97	0.001
Denitrification	14.3	9.8	-31	0.05
CO_2 Production	31	12	-61	0.001
Phosphatase	0.41	0.24	-40	0.001
Redox Potential	-107	-193	-80	0.001

	Units
Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}
% Respiration	%
Nitrogen fixation	ng x g. dry wt.$^{-1}$ x h^{-1}
Denitrification	ng x g. dry wt.$^{-1}$ x h^{-1}
CO_2 Production	nM x g. dry wt.$^{-1}$ x h^{-1}
Phosphatase activity	µM PNP x g. dry wt.$^{-1}$ x h^{-1}
Redox potential	mV

Table 35. Percent changes in variables in response to 5 month exposure to crude oil and crude oil + Corexit.

	Trays		Aquaria	
Variable	Oil	Oil + Corexit	Oil	Oil + Corexit
A. Uptake				
Glucose Uptake	-60	-84	-69	-61
Glutamate Uptake	-71	-85	-73	-76
Glutamate Vmax	-78	-90	-72	-79
Glutamate Tt	+117	+113	+223	+200
Glutamate Percent Resp.	9	+8	+14	+11
Glucose Percent Resp.	+264	+240	+133	+148
B. Total Adenylates	-74	-87	-65	-68
C. Surface Eh	-380	-336	-576	-561
D. Nitrogenase	-85	-82		

we conducted a series of experiments in which perturbed sediments were reexposed to these same pollutants during short-term challenge exposures (Table 36). The sediments which were not previously exposed to these pollutants (i.e. the controls) showed the same response that we have consistently observed in our short-term challenge experiments that we reported earlier in this section. The sediments that had been exposed to crude oil for up to 5 mo (subsection "B: of Table 36) showed little addition effect when more crude oil was added under the acute, short-term challenge conditions. In sediments exposed for 8 months or longer, there was a significant reduction in uptake rates in the sediments previously exposed to crude oil. When these sediments were exposed to crude oil and Corexit or Corexit alone at 50 ppm, there was a large reduction in uptake rates. In the sediments that had been exposed to Corexit and crude oil for up to 5 months (subsection "C" of Table 36), the resulting observations were essentially the same. These observations suggest that benthic microorganisms that have had prior exposure to Corexit, do not adapt to its presence. Either the original Corexit was utilized by the microorganisms present and/or bound to the sediments so that it was no longer affecting the population or there was a reestablishment of the a susceptable population.

The crude oil challenge experiments suggest that either the population adapted to the presence of crude oil or that whatever was causing the reduction in uptake rates in the acute challenge experiments was still present in the sediments exposed to crude oil for up to 5 months.

The changes in relative microbial activity described above were measured using the one substrate concentration method (Tables 27-31). We also measured relative microbial activity using the

Table 36. Short-term crude oil and Corexit effects on glucose uptake rates in sediments that were non-oiled and those treated with crude oil or crude oil and Corexit for periods of time greater than 1.5 months. These sediments were collected at Kasitsna Bay. The values given are the percent reduction in observed uptake rates compared to controls.

A. Sediments which were not exposed in short-term challenge experiments.

Month	Exposure time in months	Short-term effects caused by Crude Oil	Crude Oil + Corexit	Corexit
April	1.5	33	51	36
July	5	28	68	58
Nov.	8	49	-	-
Jan.	11	30	-	-

B. Sediments that were challenged with crude oil.

Month	Exposure time in months	Short-term effects caused by Crude Oil	Crude Oil + Corexit	Corexit
April	1.5	0	56	57
July	5	2	79	72
Nov.	8	25	-	-
Jan.	11	23	-	-

C. Sediments that were challenged with crude oil and Corexit.

Month	Exposure time in months	Short-term effects caused by Crude Oil	Crude Oil + Corexit	Corexit
April	1.5	4	61	58
July	5	7	71	72

multi-substrate concentration technique of Wright and Hobbie (1966). This procedure is described in detail in Section I of this report. We used this technique to measure the effects of crude oil on relative microbial activity (V_{max}) and on the time required for the natural microbial population to utilize the naturally occurring substrate available (T_t). In all cases, the relative microbial activity decreased and the T_t values increased; all of these differences were statistically significant. The transport constant plus the natural substrate concentration ($K_t + S_n$) value was also measured but this value was not consistently affected.

2. Effects of crude oil on respiration percentages by benthic microorganisms.

During our studies, we measured the effects of crude oil on respiration percentages. A summary of the results from the initial experiment are reported in Tables 27-31. In all cases, the mean percent respiration was higher in sediments exposed to crude oil than those that were not regardless of the labeled substrate used to make that determination. In general, the increase in the percent respiration in treated sediments was higher when glucose was used as the test substrate than when glutamic acid was used. The amount of change in percent respiration was about the same in the first set of observations (Table 27) as in the 11 month observations (Table 30); however, after the sediments had been exposed for 18 months, the change in the percent respiration was reduced (Table 31). The percent respiration also increased in sediments exposed to "weathered" crude oil (Table 32) and in untreated sediments overlayed with crude oil treated sediments (Table 33). The percent respiration was also repressed in treated Coal Bay sediments (Table 34).

In a series of three studies, we observed the effects of various concentrations of fresh crude oil on respiration percentages in three sediments (Fig. 49). Significant changes were observed at concentrations at or below 1 ppt. The change in percent respiration suggested that basic metabolic and/or population changes had occurred in the treated sediments. When the radioactivity associated with the cells and the CO_2 fractions are plotted separately (Fig. 50), it becomes apparent that the presence of crude oil has very little effect on CO_2 production rates from the labeled substrate used (glucose in this case); however, the rate at which the substrate is incorporated into cell material is greatly affected. This suggests that crude oil interferes with biosynthesis and thus the production of microbial biomass.

3. The Elson Lagoon study.

In the Elson Lagoon study we observed effects of crude oil on relative microbial activity in sediments. The results were similar to those observed during the course of the Kasitsna Bay study. The main difference was that instead of taking as little as 5 weeks to observe an effect, as it did in Kasitsna Bay, it took 9-12 months of exposure before measurable differences were observed (Table 37). Once the effects of crude oil on microbial activity were initiated, they were as great as those found in the Kasitsna Bay sediments. Reduced microbial activity was observed in sediments that had been exposed to crude oil for up to 2 years (the last observation in this series). We also observed increases in the respiration percentages

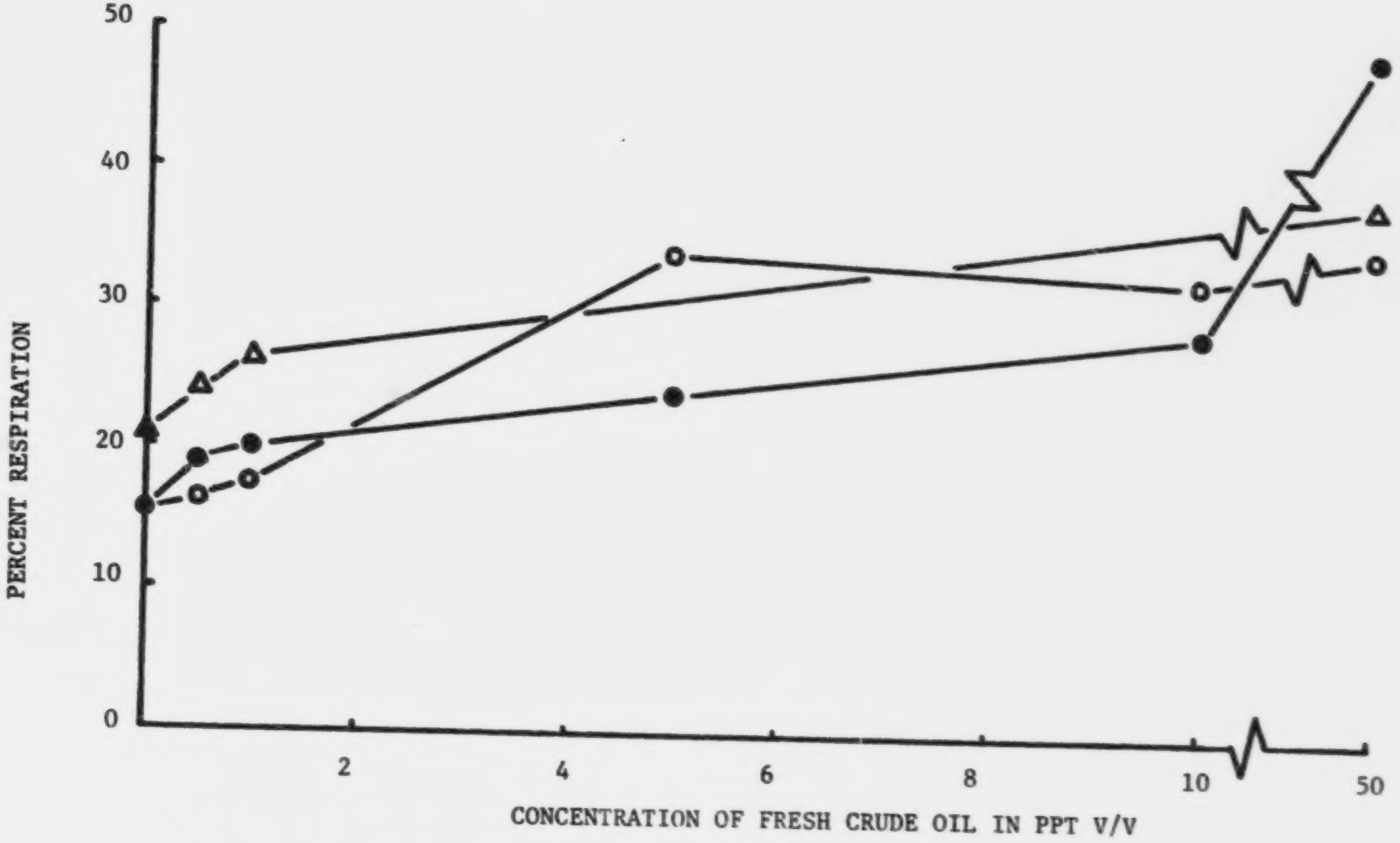

Figure 49. Effects of various concentrations of crude oil on respiration percentages in three marine sediments.

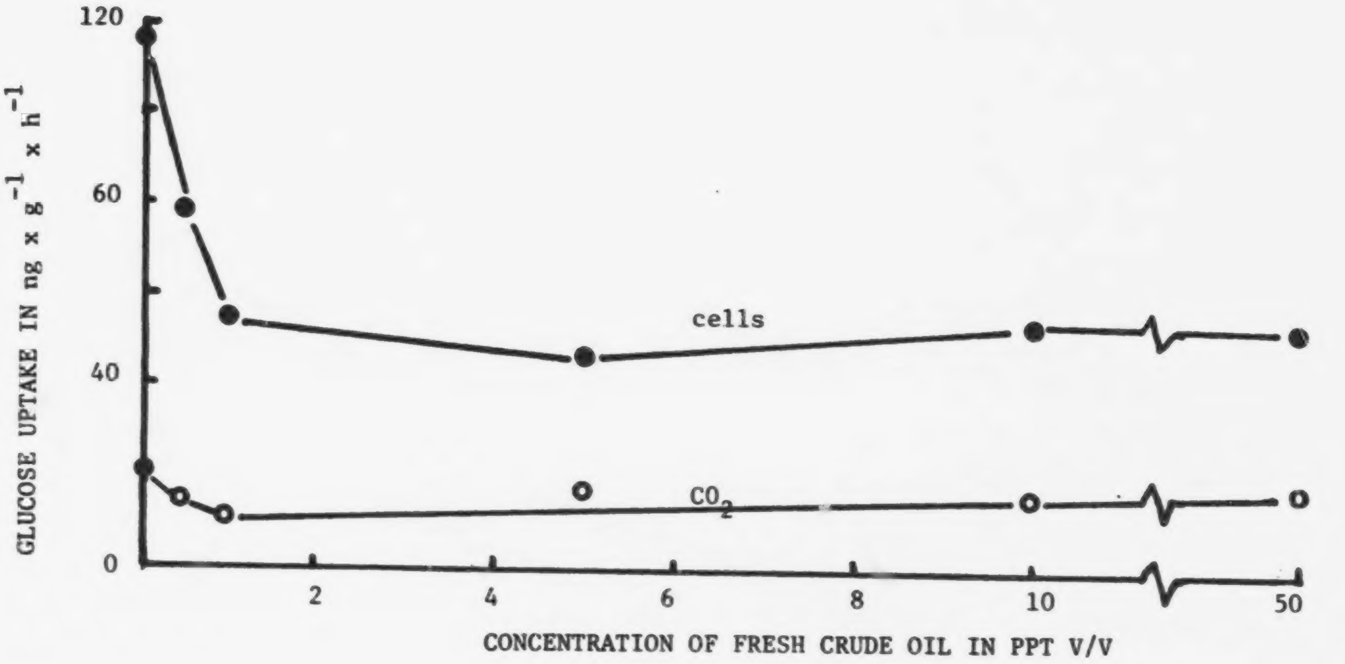

Figure 50. Effects of various concentrations of crude oil on incorporation of glucose into cell material and respiration to CO$_2$ after an exposure of 3 mo.

Table 37. Long-term effects of crude oil on glucose and glutamate uptake in the Elson Lagoon sediments.

		Uptake				
		Glucose		Glutamate		
Date	Exposure	uptake[1]	Δ%[2]	uptake[1]	Δ%[2]	Vmax[3]
Jan. 1978	Control	3.4		23		0.14
	"	0.2		3		0.25
	"	1.2		6		-
	Oil-6 months	5.7	NS	44	NS	0.09
	"	7.9		15		0.10
	"	2.0		15		-
Apr. 1978	Control	12.2		479		3.5
	Oil-3 months	3.2	NS	393	NS	8.2
Aug. 1978	Control	2.7		19		-
	"	13.2		161		-
	Oil-8 months	6.9	NS	96	NS	-
	Oil-4 months	1.5		9		-
Jan. 1979	Control	2.4		7		-
	"	3.7		8		-
	Oil-12 months	0.8	-74	2	-73	-
	Oil-9 months	0.8	-74	3	-60	-
Aug. 1979	Control	172		1025		0.8
	Oil-20 months	25	-85	152	-85	0.1
	Oil-16 months	38	-72	283	-72	0.2
Jan. 1980	Control	12		208		-
	"	32		75		-
	Oil-24 months	8	-64	75	NS	-

[1] ng/g dry wt/hr.

[2] Percent change relative to control; NS - not significant if ranges overlap.

[3] µg/g dry wt/hr.

in all oiled sediments analyzed (Table 38). These changes were much lower than those observed in the Kasitsna Bay sediments. Even though we observed an increase in respiration percentages in each case, these differences were not statistically significant.

4. The Glacier Bay study.

In June 1979, a cruise ship spilled fuel oil into Glacier Bay near Juneau, AK. In October 1979, we collected three oiled and three non-oiled sediments for analysis at the Kasitsna Bay laboratory. Care was taken to collect non-oiled sediments which approximated the physical characteristics of the oiled samples. We conducted a series of uptake experiments on these sediments using glucose to determine the effects of the oil on relative microbial activity and respiration percentages (Table 39). We then compared the resulting measurements on a sample by sample basis; matching oiled and non-oiled sediments with the same physical characteristics. If the microbial populations in the exposed sediments were affected by the presence of the crude oil, we would expect to find reduced relative microbial activity, increased respiration percentages and reduced effects on relative microbial activity in acute fresh crude oil challenge experiments. In 2 out of 3 samples, the relative microbial activity was reduced and the percent respiration was higher. In all three samples, there was a lower reduction in microbial activity in the oiled samples when the samples challenged with fresh crude oil during the assay.

5. Long-term crude oil effects on nitrogen fixation rates.

The effects of crude oil on nitrogen fixation was one of the variables that we assayed during the initial long-term crude

Table 38. Long-term effects of crude oil on percent respiration in Elson Lagoon sediments.

	Exposure	% Resp. Glucose	Δ%	% Resp. Glutamate	Δ%
Jan 1978	Control	25		44	
	Control	20		41	
	Control	23		51	
	Oil 6 months	20	+9	45	+4
	" "	27		49	
	" "	27		48	
Apr. 1978	Control	19		44	
	Oil 3 months	29	+53	62	+41
Aug. 1978	Control	29		54	
	Control	-		46	
	Oil 8 months	23	-21	-	
	Oil 4 months	57	+97	54	+8
Jan. 1979	Control	49		78	
	Control	47		7.4	
	Oil 12 months	71	+48	90	+18
	Oil 9 months	59	+23	81	+ 7
Aug. 1979	Control	20		48	
	Oil 20 months	38	+90	49	+ 2
	Oil 16 months	33	+65	53	+10
Jan. 1980	Control	32		53	
	Control	23		46	
	Oil 2 years	32	+16	63	+12

Table 39. Effects of fuel oil on relative microbial activity and respiration percentages in Glacier Bay, AK sediments. Each sample was exposed to crude oil in a short-term challenge experiment.

Sediment type			Glucose Uptake		Percent change	Percent respiration
			no oil	oil		
A.	Coarse sand	Non-oiled	138	42	-70	23
		Oiled	156	83	-47	47
			+13*			+104*
B.	Sand-silt	Non-oiled	30	15	-47	17
		Oiled	18	11	-39	30
			-40*			+76*
C.	Glacial flour	Non-oiled	6.8	5.3	-22	36
		Oiled	2.4	7.1	198	29
			-65*			-26*

1 = Glucose uptake in units of ng x g^{-1} dry wt. x h^{-1}.

* = These values are the percent change relative to the controls.

oil exposure experiment. Whenever we made this observation, there was a very significant reduction in nitrogen fixation rates (Tables 27-31). This was only observed when fresh crude oil was used. When the sediments were exposed to "weathered" crude oil, there was no reduction in nitrogen fixation rates (Tables 32 and 33). There was a reduction in nitrogen fixation rates when fresh crude oil was applied as an overlay (Table 33) or when sediments from another location in Cook Inlet were used; i.e. the Coal Bay experiment listed in Table 34.

We also wanted to determine the effect of various concentrations of fresh crude oil on nitrogen fix rates and to determine how long it took for these alterations to take place. In one series of experiments, we observed changes in nitrogen fixation rates in sediments exposed for varying lengths of time to various concentrations of fresh crude oil (Fig. 51). In all three sediment samples, there was approximately a 50% reduction in nitrogen fixation rate when the crude oil concentration was 1 ppt. When the concentrations were increased up to 50 ppt, there was a further reduction in rates observed. At this concentration, the rate was only about 20% of that found in the control (nontreated sediment).

During another set of experiments, we observed the length of time required for nitrogen fixation rates to be altered in the presence of 50 ppt crude oil (Fig. 52). The experiment was conducted on three sediments of varying characteristics which were collected from three different locations. In all three sediments, there was a marked reduction in nitrogen fixation rates after the sediments had been exposed for 5.5 days. Measurable reductions were also observed after the sediments had been exposed for only 1.5 days.

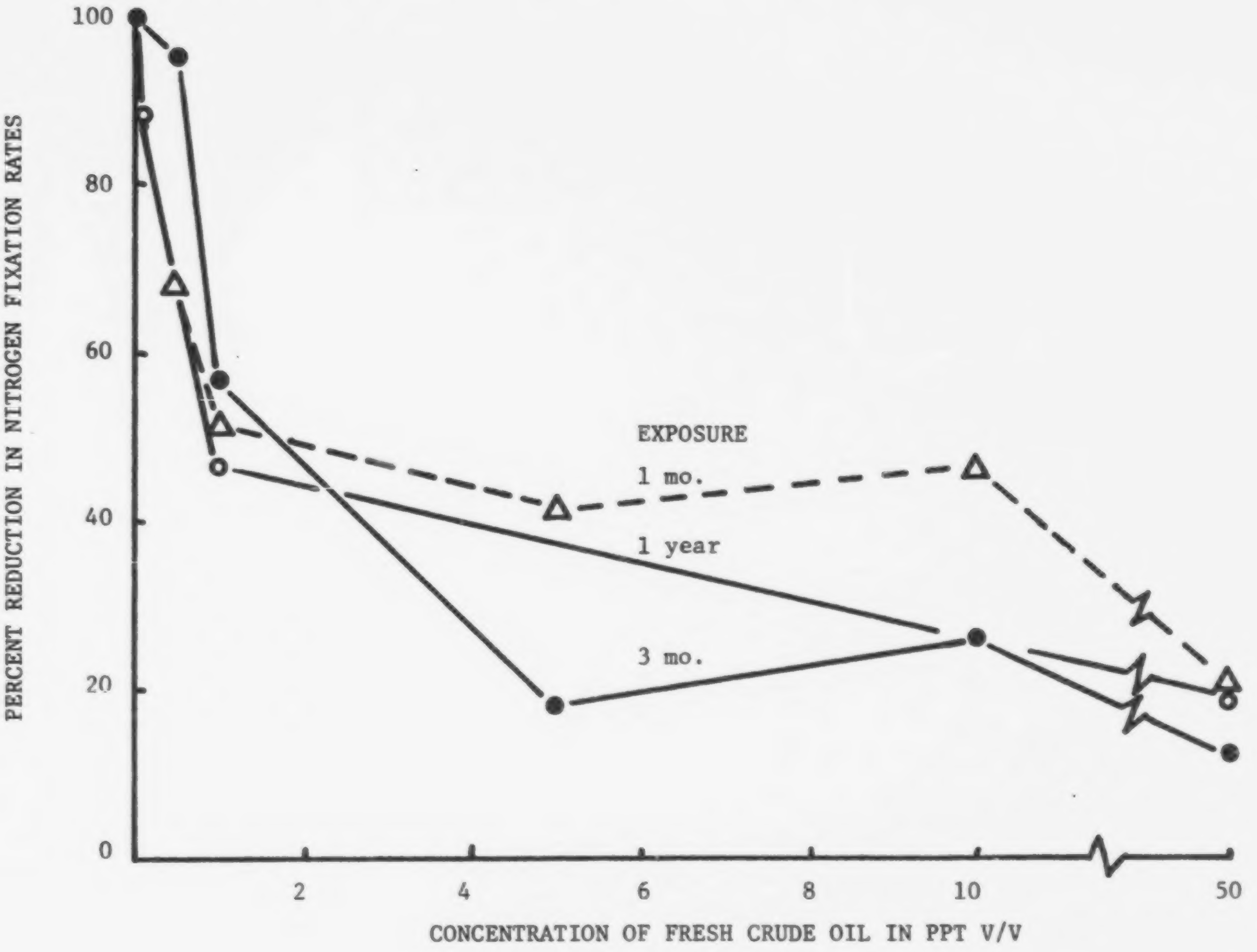

Figure 51. Effects of various concentrations of fresh crude oil on nitrogen fixation rates in three marine sediments expressed as a percent reduction relative to controles.

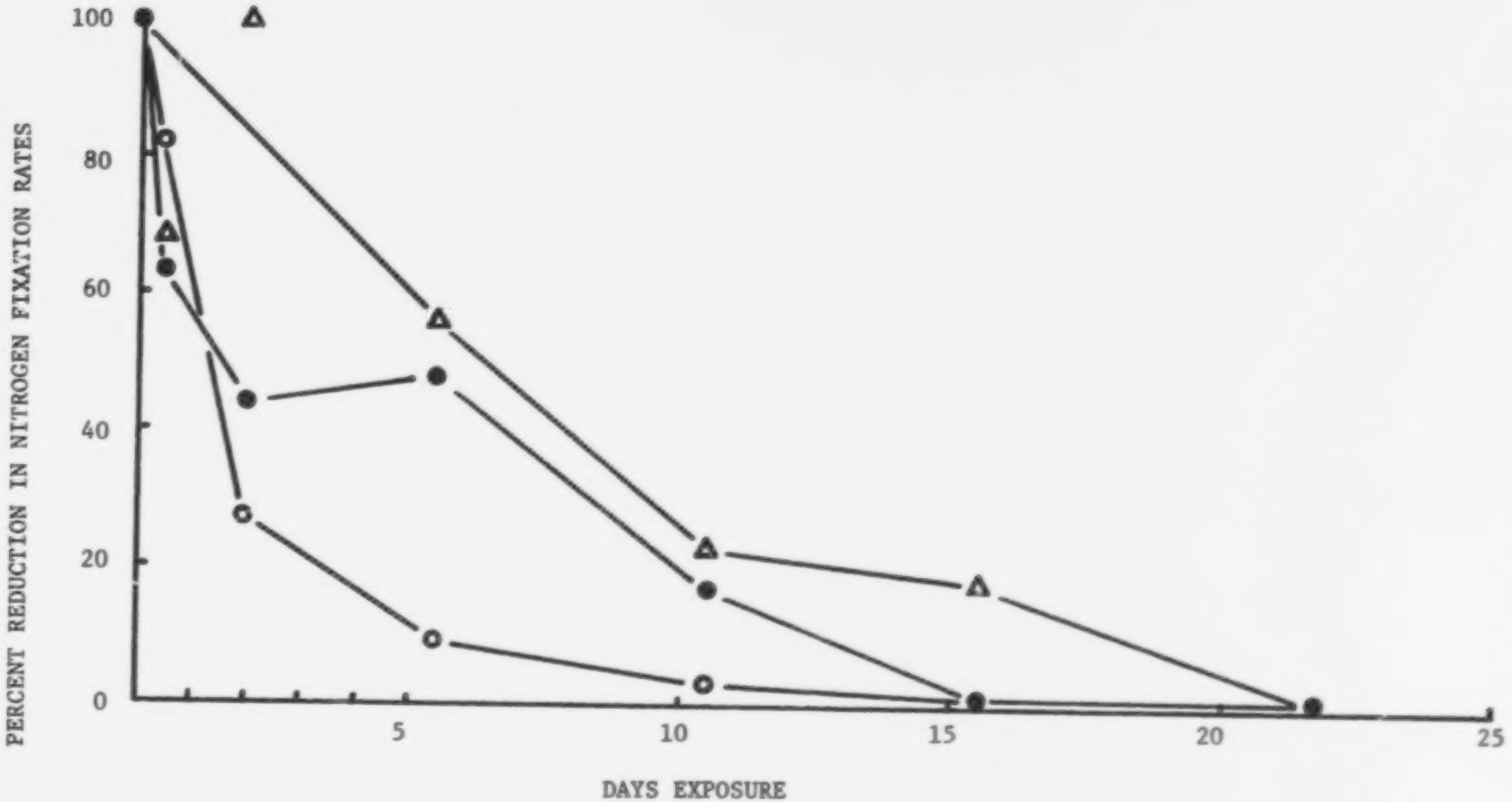

Figure 52. Effects of fresh crude oil at 50 ppt on nitrogen fixation rates in three marine sediments exposed for various lengths of time.

We also studied the effects of fresh Prudhoe Bay crude oil on nitrogen fixation rates in sediments collected in Elson Lagoon (Beaufort Sea). Even after exposure times of up to 8 mo, we did not observe measurable reductions in nitrogen fixation rates (Table 40).

6. Long-term effects of crude oil denitficiation rates.

In the initial long-term crude oil effects study conducted at Kasitsna Bay, we started to monitor effects on denitrifcation potentials after the sediments had been exposed to crude oil for 8 mo (Table 29). At this and subsequent field periods when these measurements were made, we observed a significant reduction in denitrification potentials. Significant reductions were also observed in sediments exposed to "weathered" crude oil (Tables 32 and 33).

During the January study, both natural and potential denitrification rates were measured (Table 30). Natural denitrification rates in oiled sediments were reduced below the level of detection (approximately 0.003 ng N_2O). When the potential denitrification rates were measured, they were found to be approximately 1,000 times the natural rate. Crude oil exposure reduced denitrification rates by 74% under those conditions. The significance of the differences between the means was $p<0.004$ for both natural and potential rates.

As in the relative microbial activity and nitrogen fixation studies, we measured the effects of various concentrations of fresh crude oil on denitrification potentials (Fig. 53). In the sediment that was exposed to crude oil for 4.5 weeks, there was very little effect noted regardless of the concentration of oil used. In the sediments that had been exposed for 3 mo, there was not a dramatic

Table 40. Long-term effects of crude oil on nitrogen fixation rates in Elson Lagoon.

	Exposure time	Control[1]	Oil[1]
Apr. 1978	0	1.3	1.2
Apr. 1978	3 mo.	1.4	1.9
Aug. 1978	1 day	0.3	0.4
Aug. 1978	4 mo.	0.3	0.4
Aug. 1978	8 mo.	0.3	0.4

[1] ngN_2 fixed x g dry $wt.^{-1}$ x $hr.^{-1}$.

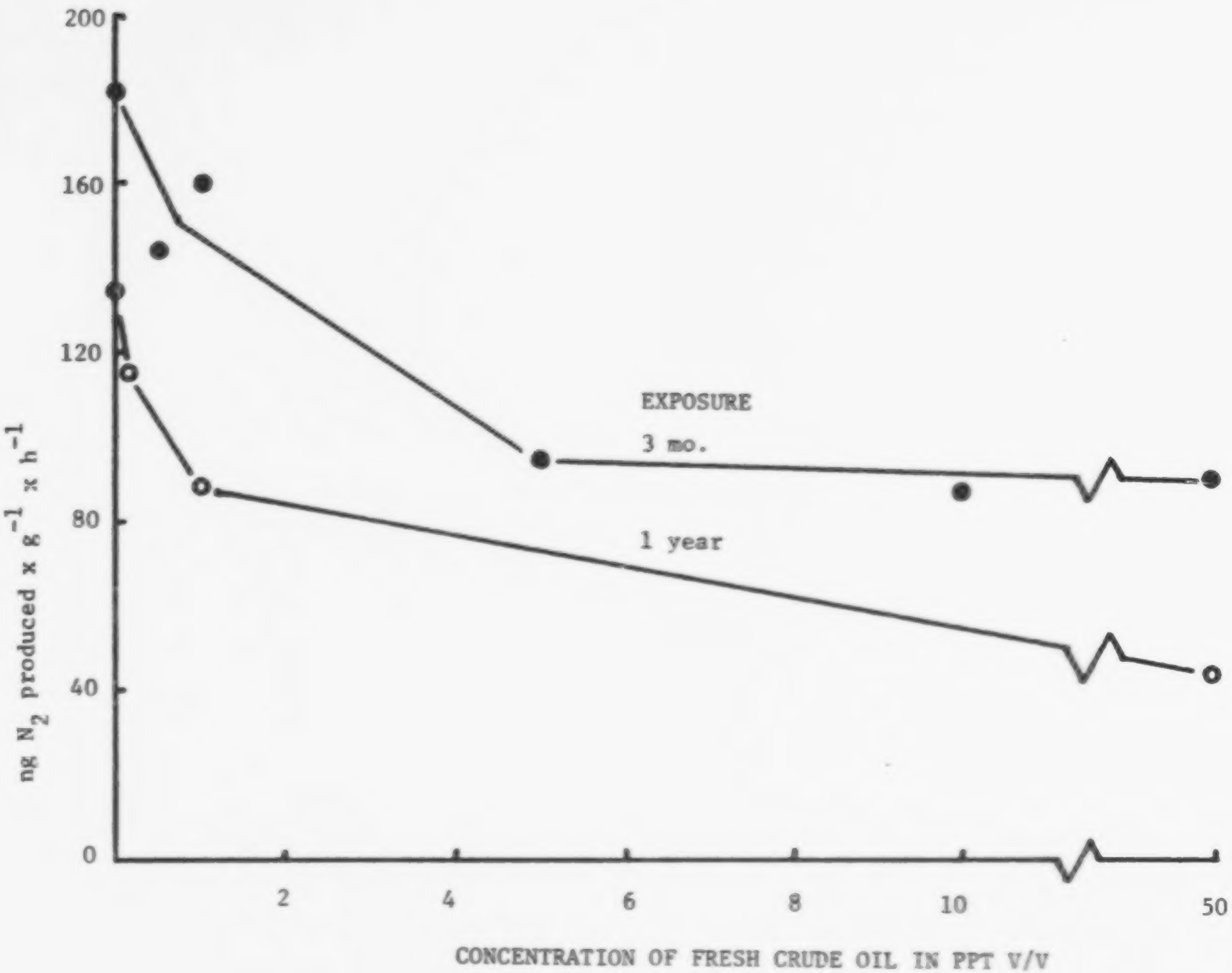

Figure 53. Effects of fresh crude oil on denitrification rates in marine sediments to which nitrate had been added.

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reduction in denitrification at concentrations below 5 ppt. In the sediments that had been exposed for one year; however, there was a marked reduction at 1 ppt.

We also conducted an experiment to determine how long it takes for a significant reduction in denitrification potentials to occur in sediments exposed to fresh crude oil at 50 ppt (Fig. 54). In two of the three sediments tested, there was little effect during the 22 day exposure period. In the third sediment, there was a large drop in denitrification activity at the beginning of the study but very little net change after that time.

7. Long-term crude oil effects on CO_2 and methane evolution rates.

During the course of the initial long-term exposure experiment conducted at Kasitsna Bay, we also observed that the CO_2 and methane evolution rates were higher in the oiled sediments than in nonoiled sediments (Tables 20 to 31). This was not the case however, in the sediment sample studied from Coal Bay (Table 34). In this sample there was a significant reduction in the CO_2 production rate (methane evolution rates were not determined). The exposure time in this experiment was 8 mo.

In addition to these observations, we also conducted a series of three experiments in which the effects of various concentrations of crude oil on CO_2 evolution rates was observed (Fig. 55). In the sediment that was exposed for 1 day, there was a reduction in the rate of CO_2 evolution observed at a concentration of 1 ppt or higher. In the sediments that had been exposed for 4.5 weeks or longer, there was an increase in CO_2 evolution rates observed at concentrations of 1 ppt or greater. When a time course experiment was conducted on three sediments exposed to 50 ppt crude oil, we observed an initial

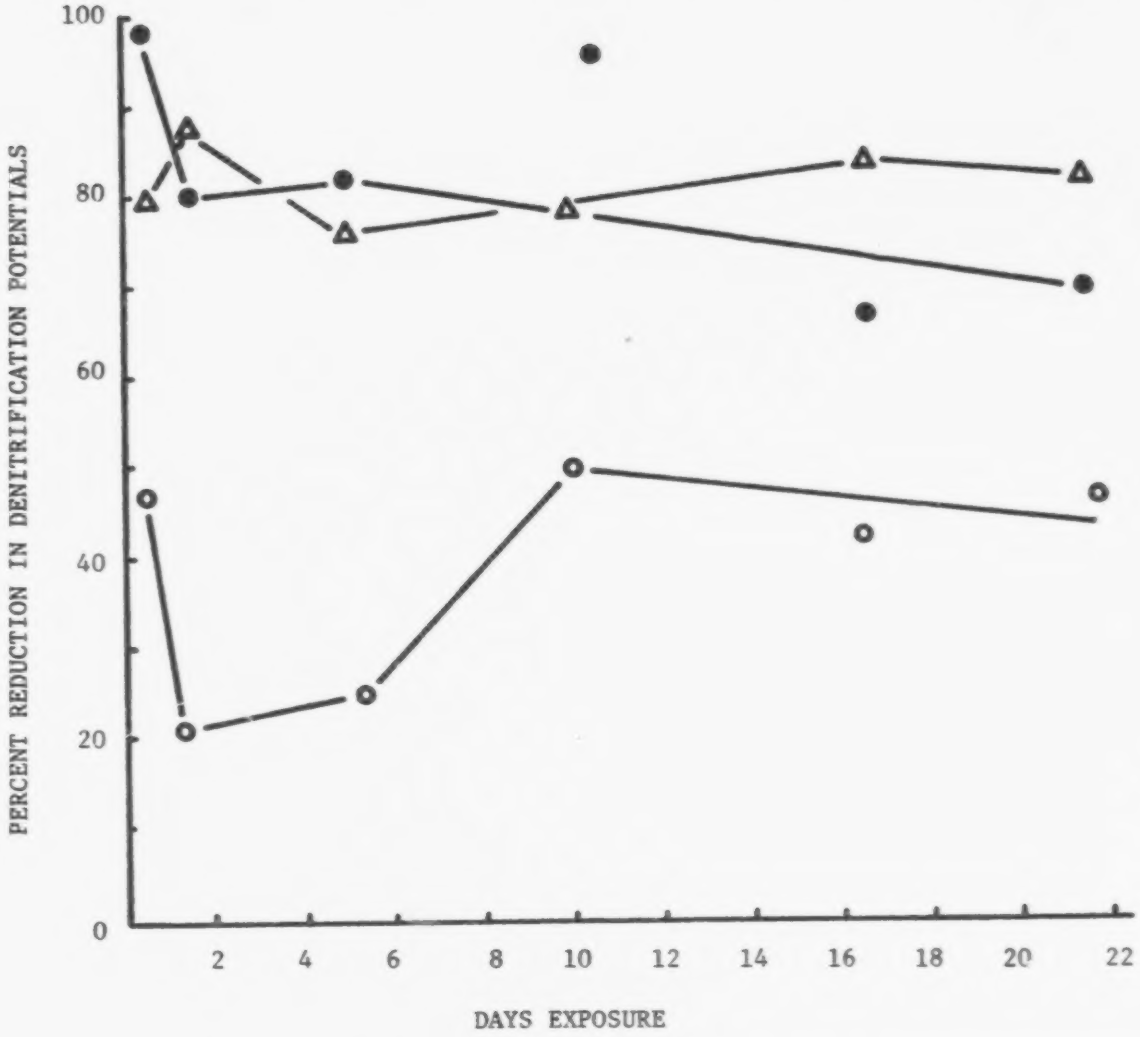

Figure 54. Effects of fresh crude oil at 50 ppt on denitrification potentials in three marine sediment samples.

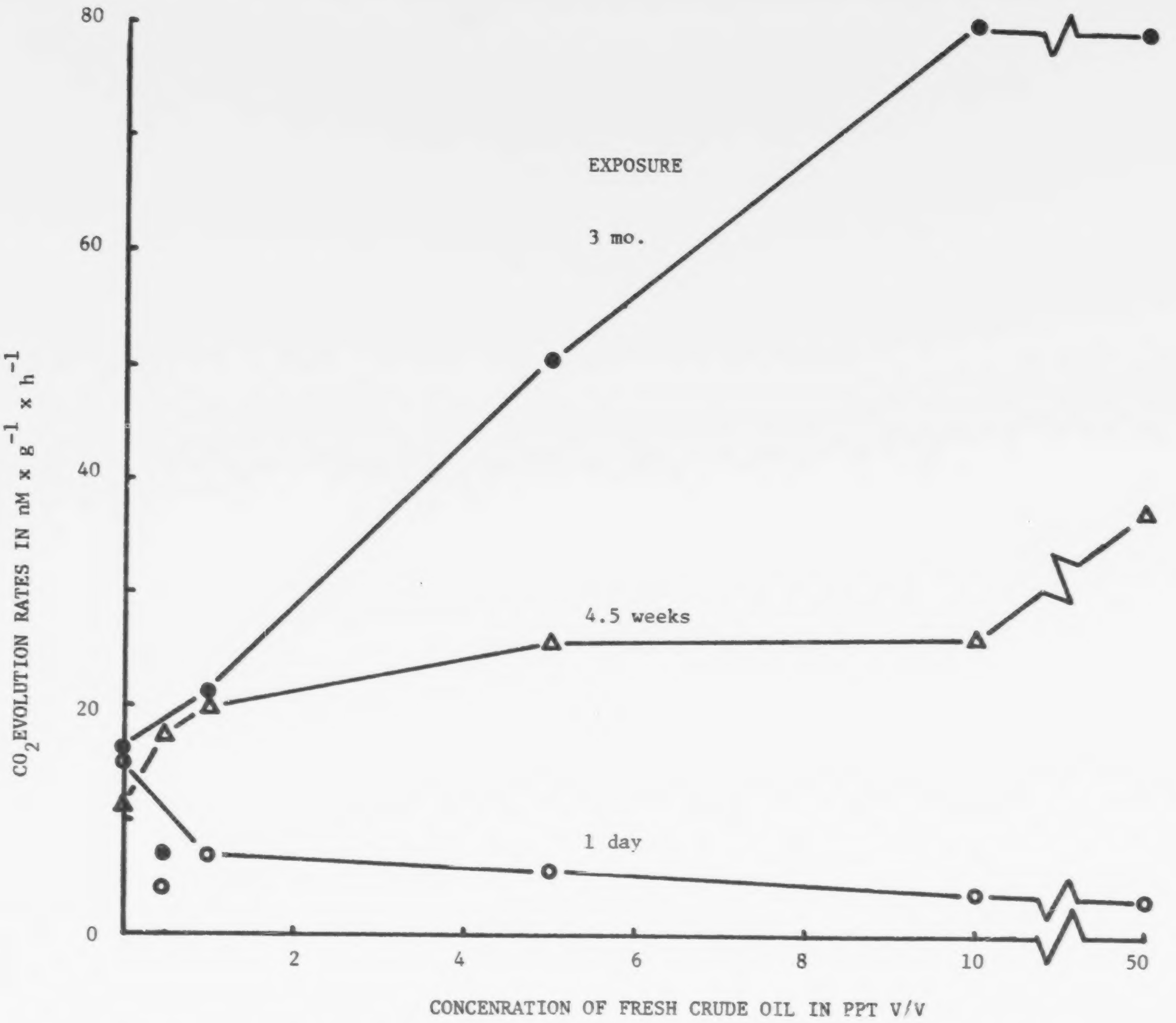

Figure 55. Effects of fresh crude oil on CO_2 production in three marine sediments.

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depression in CO_2 evolution rates (Fig. 56). When the sediments were exposed to crude oil for about 10 to 16 days, the rates became higher than the controls.

During our long-term crude oil exposure experiments, we also observed elevated methane concentrations in the treated sediments (Tables 30,31,32,33).

When the effects of crude oil at various concentrations on methane concentration was measured, we found that a measurable increase could be detected after 20 days exposure at a concentration as low as 0.5 ppt (Fig. 57). When a time course experiment was conducted, we observed an initial depression in methane production in two of the three sediments examined (Fig. 58). After 16 days exposure all three sediments showed elevated methane production rates.

8. Long-term oil effects on total adenylates and energy charge.

During two field periods in the Elson Lagoon study, we measured the concentrations of ATP, ADP, and AMP in oiled and nonoiled sediments (Table 41). The concentration of total adenylates is calculated by adding the individual concentrations of all three adenylate species. In all sediments that had been exposed to crude oil for 4 months or more, the concentration of total adenylates was significantly lower in the oiled samples. The same measurements were made in Kasitsna Bay sediments during two field periods (Tables 27 and 28). The same phenomenon was observed in these sediments as well.

Although there was a large seasonal change in the energy charge calculated from the adenylate concentrations in the Elson Lagoon samples, there was no significant difference between oiled

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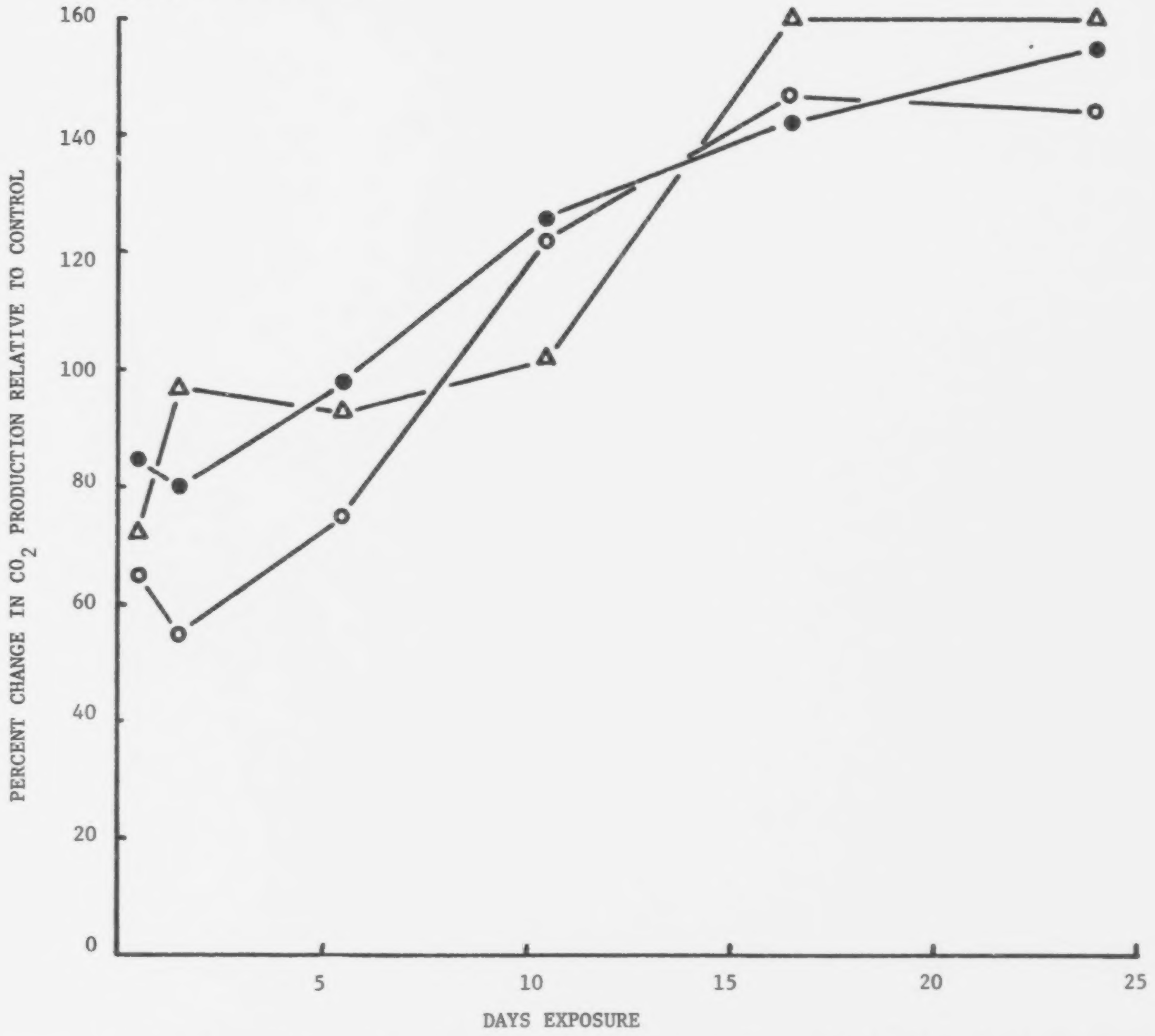

Figure 56. Effects of fresh crude oil at 50 ppt on CO_2 evolution from three marine sediments.

634

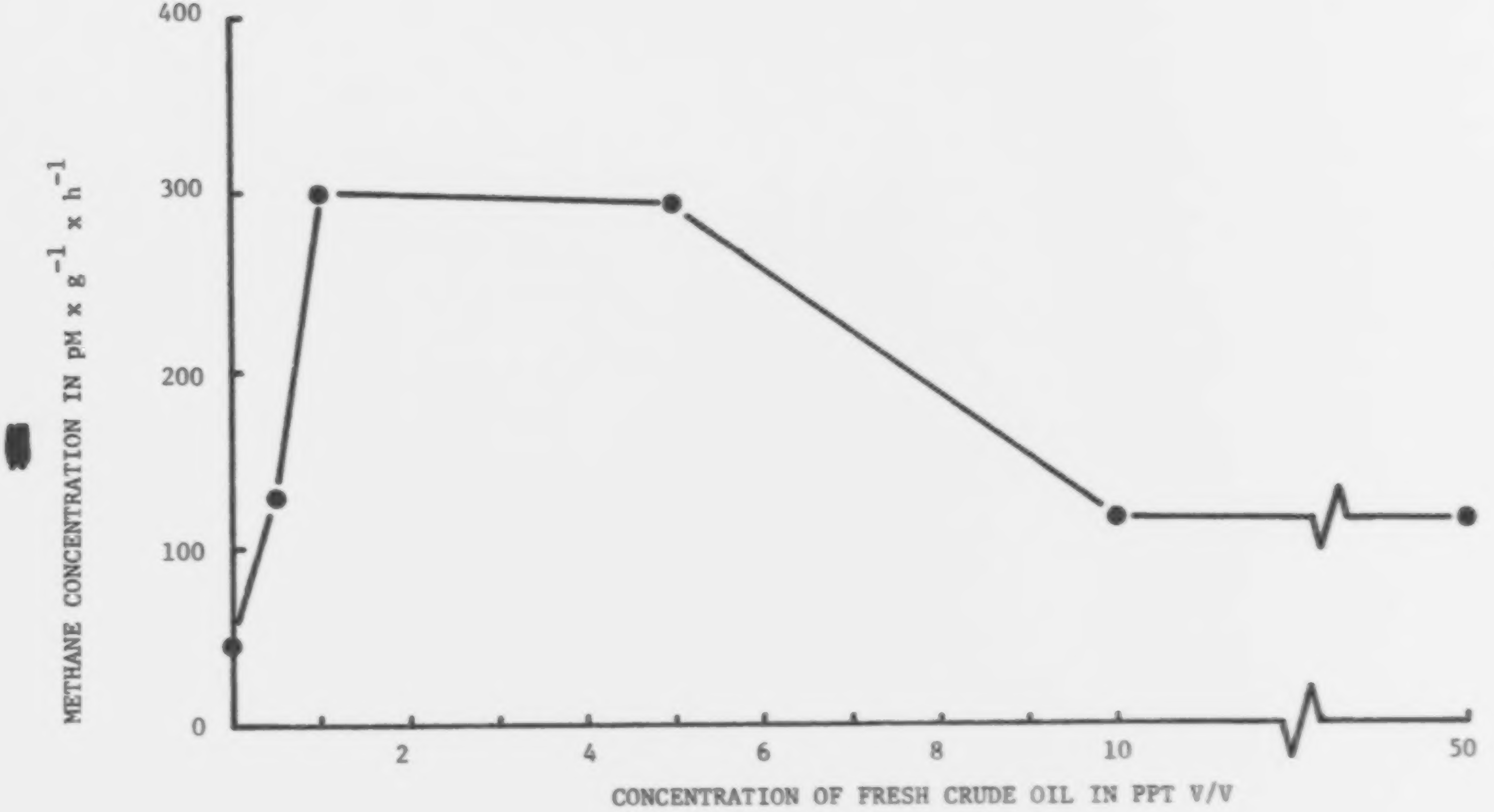

Figure 57. Effects of crude oil at various concentrations on methane concentration in a marine sediment exposed for 20 days.

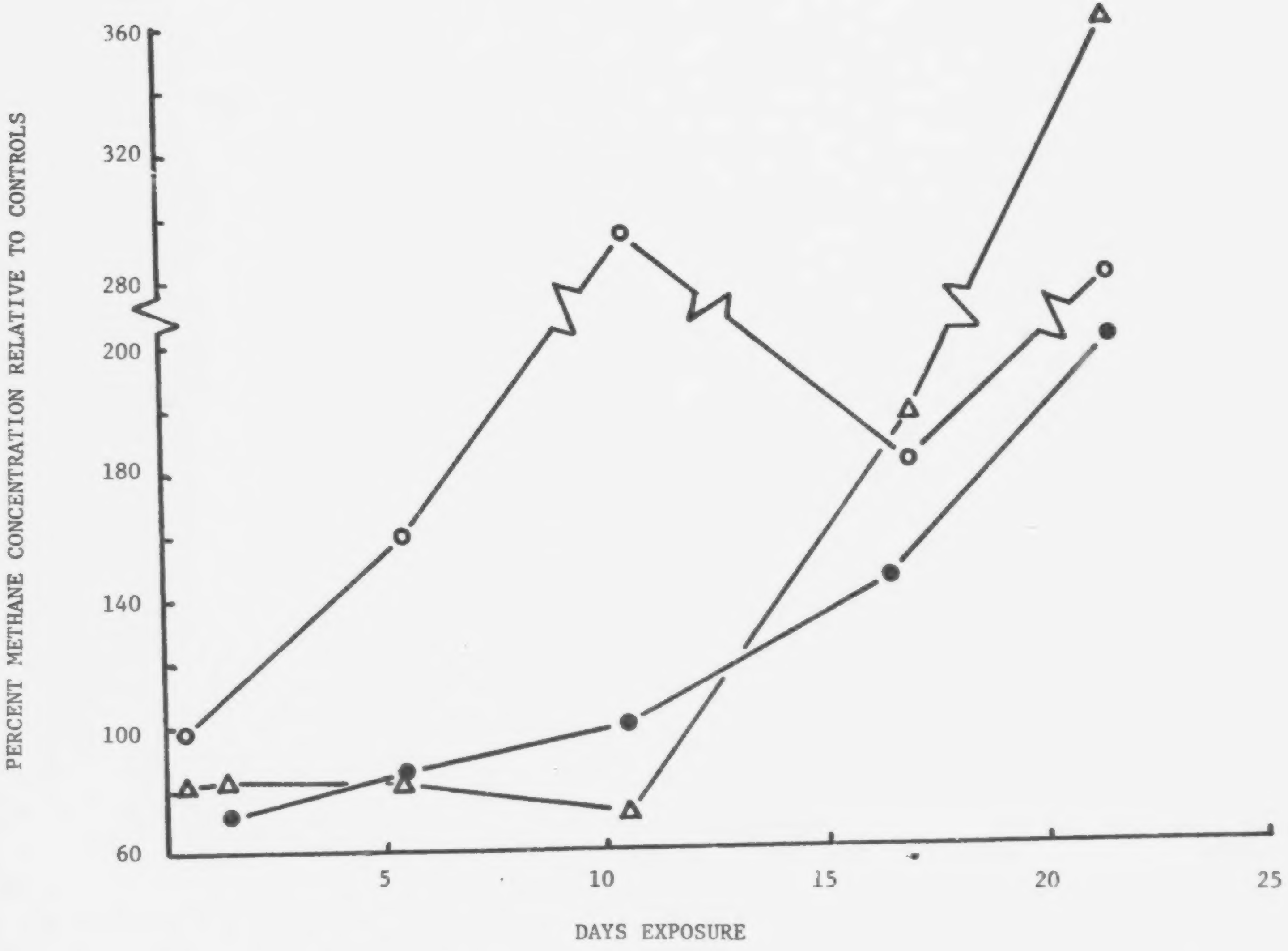

Figure 58. Effects of fresh crude oil at 50 ppt on methane concentration in three marine sediments exposed for varying lengths of time.

Table 41. Adenylate measurements made on sediment samples collected during the summer, 1978 Beaufort Sea study and in the Elson Lagoon in January, 1979.

A. Beaufort Sea, August, 1978

Sample Number	Treatment	*ATP	*ADP	*AMP	Total Adenylate	Energy Charge
BB601	Control	245	6	7	263	0.95
BB602	Oiled 4 mo	3.2	0	0	3.2	1.00
BB605	Oiled 8 mo	3.6	0.2	0	3.7	0.98
BB607	Control	211	19	0	227	0.96
BB619	Offshore	337	7	10	352	0.96
BB624	Offshore	13.7	1.3	0.6	15.7	0.92
BB262	Offshore	23.7	0.3	0	24.6	0.99
BB627	Offshore	62.7	1.9	0.3	65.3	0.98

B. Elson Lagoon, January 1979

Sample Number	Treatment	*ATP	*ADP	*AMP	Total Adenylate	Energy Charge
BB701	Oiled 9 mo.	0.2	0.08	0.28	0.4	0.24
BB702	Oiled 12 mo	0.33	0.54	1.01	1.9	0.38
BB703	Control	0.28	4.0	13	17	0.25
BB703	+ oil	0.63	5.4	17	23	0.27
BB704	Control	0.07	1.9	11	13	0.15
BB705	Oil 0 time	0.17	3.0	11	14	0.19

*nMoles of adenylate/g dry wt.

and nonoiled sediments. In both Kasitsna Bay studies, the mean energy charge values were lower in the oiled samples. However, the difference was statistically significant in only one of the two studies.

9. Long-term crude oil effects on bacterial concentrations.

During the April, July and November 1979 and July 1980 Kasitsna Bay field periods, we measured the concentrations of bacteria in oiled and nonoiled sediments using a direct counting technique (epifluorescent microscopy). In the first studies we observed mean value differences representing a 33% and a 45% reduction in bacterial cell concentrations in the oiled sediments for the April and July 1979 studies respectively (Table 42). These differences were statistically significant. During the November and July 1980 studies there were no significant differences seen between the mean values.

10. Long-term crude oil effects on sediment surface pH and Eh.

The effect of crude oil on sediment surface pH and Eh was also measured during the Kasitsna Bay study. After the sediments had been exposed to crude oil up to 5 months, there was a significant reduction in the redox potential and an increase in the hydrogen ion concentration in the surface of sediments (Tables 27 and 28). After 8 months exposure, there was still an increase in the hydrogen ion concentration but this increase was not statistically significant (Table 28). When this same variable was measured in the same sediments after 18 months exposure, there was no detectable difference in pH values when treated and nontreated sediments were compared.

This was not the case when the redox potentials were determined in the same sediments. Even after 18 months exposure, there was

Table 42. Direct bacterial concentrations in treated and non-treated Kasitsna Bay sediments.

			Cells (x 10^9)			
			Control		Oil	
Date	Source	Exposure to oil	mean	range	mean	range
Apr. 1979	Trays	1.5 months	3.9	3.2-4.6	2.5	0.6-3.8
	Aquaria	1.5 months	3.2	3.1-3.3	2.4	1.7-3.4
Jul. 1979	Trays	5 months	3.9	1.6-6.4	2.2	1.8-2.4
	Aquaria	5 months	3.7	-	1.9	1.5-2.4
Nov. 1979	Trays	8 months	3.6	2.5-4.5	3.5	2.7-4.4
Jul. 1980	Trays	18 months	0.6	.4-.8	0.5	0.3-0.6

a significant reduction in the redox potentials observed in the oiled sediments (Table 31).

In one study, we observed the effects of various concentrations of both fresh and "weathered" crude oil on redox potentials (Fig. 59). We observed measurable reductions in redox potentials in treated sediments at a depth of 8 cm at an oil concentration of 1 ppt or greater. This was true in both the sediments that had been exposed to fresh or "weathered" crude oil. A reduction in redox potential was also observed in sediments exposed to fresh crude oil for either 21 days or 3 months.

11. Long-term effects of crude oil on sediments that had been augmented with organic carbon and long-term effects on enzyme activities.

When the sediments that had been exposed to 50 ppt fresh crude oil for 1.5 years were examined for phosphatase and arylsulfatase activity (Table 31) it was found that the activity of both of these enzymes was significantly reduced ($p<0.001$). We also examined the effects of fresh crude oil at various concentrations on phosphatase activity in sediments that had been exposed for 3 months (Fig. 60). Measurable reductions were observed at a concentration of 5 ppt in these sediments; the greatest reduction was observed at 50 ppt.

The activities of these and other enzymes as well as most of the other variables measured during the course of our Kasitsna Bay studies were analyzed in a series of experiments in which sediments were augmented with various organic compounds. In these studies, replicate sets were analyzed; one set was not treated (controls) and one set was treated with 50 ppt fresh crude oil. The first set of observations (Tables 43 A, 43 B and 44) were made on two pilot

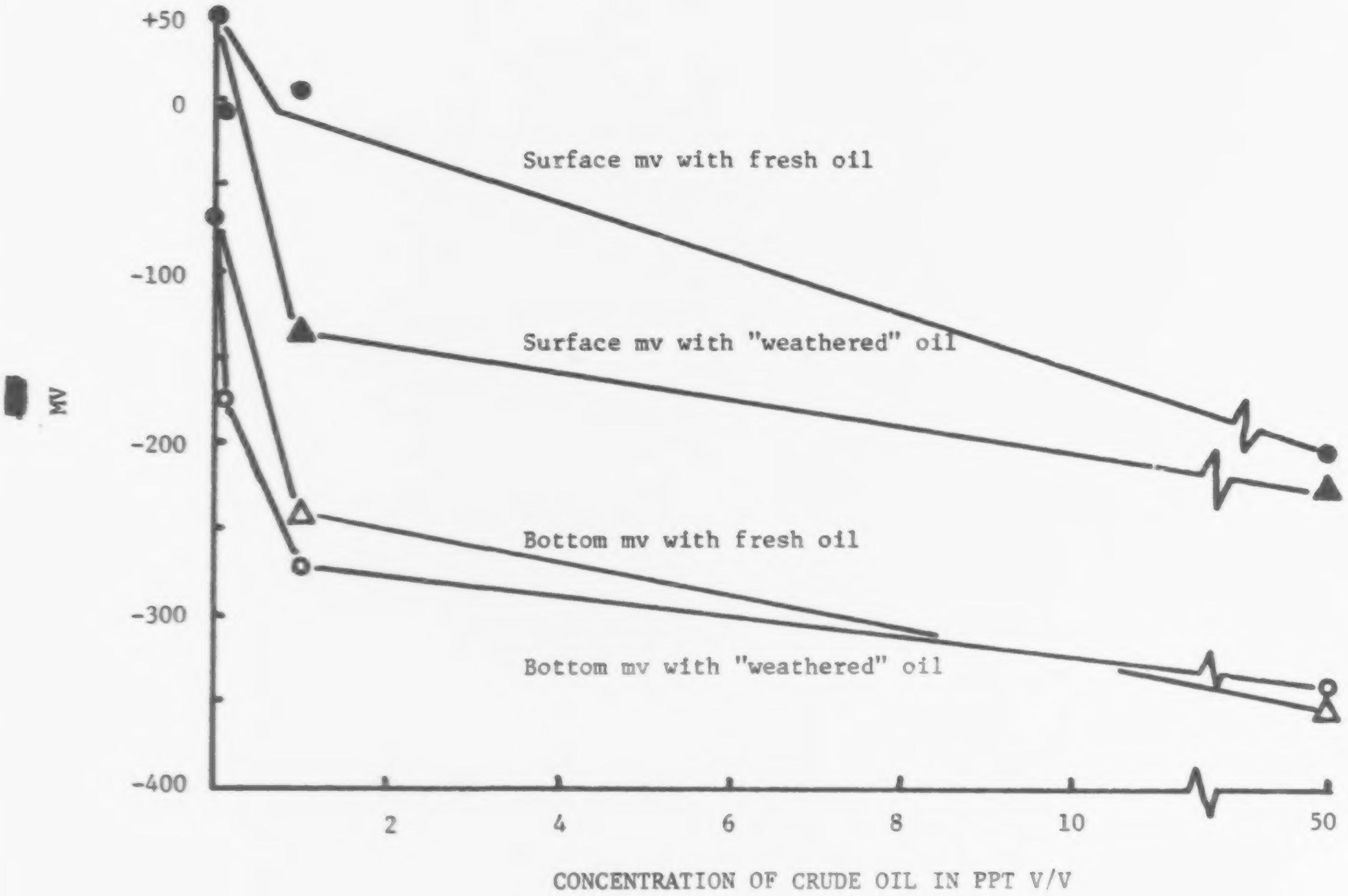

Figure 59. Effects of crude oil at various concentrations on redox potentials in a marine sediment exposed to crude oil for 1 year. Measurements were made on the surface of the sediment and near the bottom at a depth of ca. 8 cm.

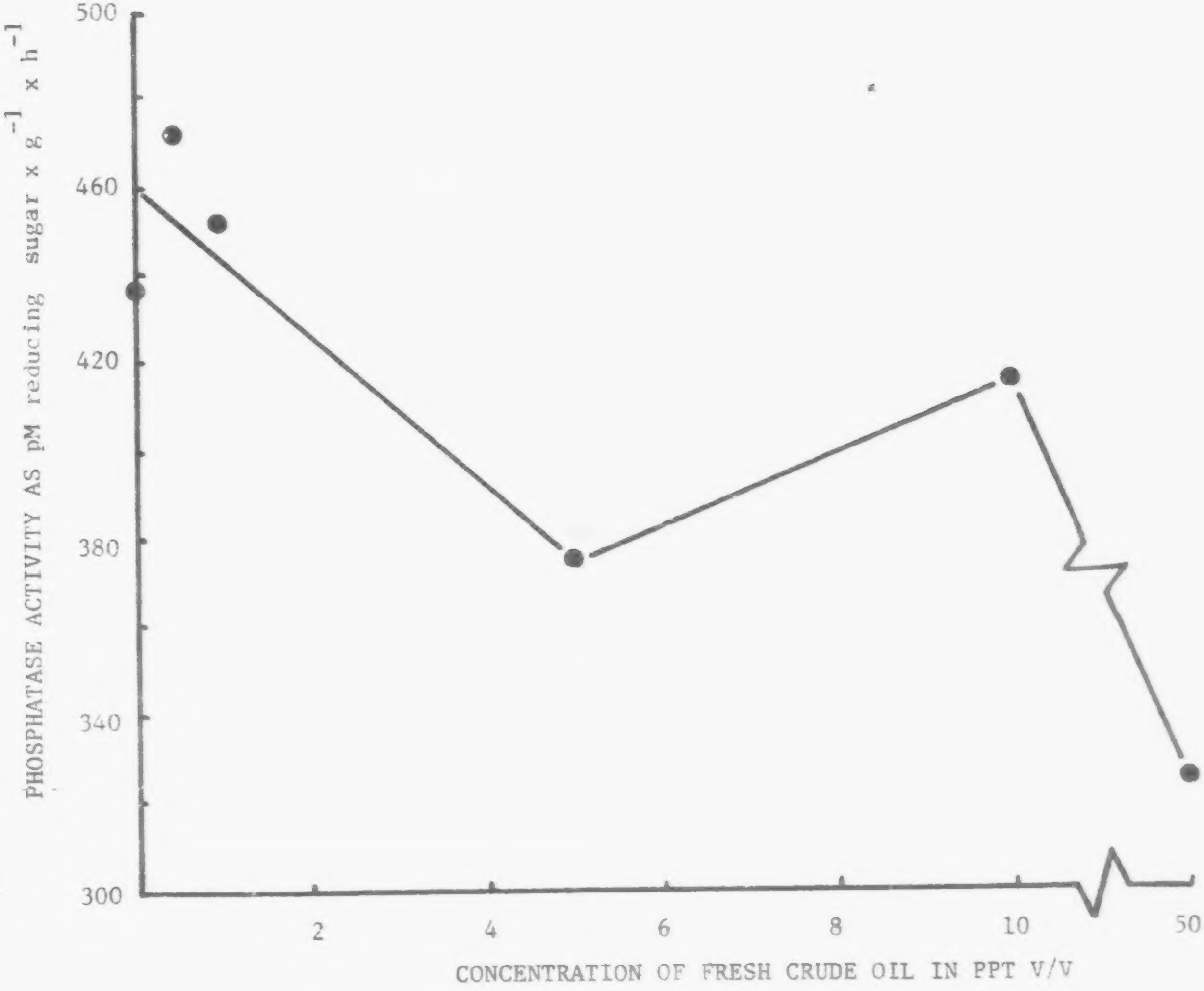

Figure 60. Effects of crude oil on phosphatase activity in a marine sediment exposed for 3 mo.

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Table 43 A. The effects of fresh crude oil at 50 ppt on various microbial processes in sediments amended with Cerophyl, chitin, or starch after 8 mo. exposure.

	CONTROL				OILED			
Variable	Control	Cerophyl	Chitin	Starch	Control	Cerophyl	Chitin	Starch
CO_2 Evolut.	0.9	8.7	5.2	2.4	3.4	15.0	15.9	1.1
% Change					+278	+72	+206	-79
N_2 Fixat.	3.1	0.5	0.5	53	0.2	0	0	5.1
% Change					-98	-100	-100	-90
Denitrif.	78	17	20	6	36	17	6	0
% Change					-54	0	-71	-100

% change compares oiled + nonoiled sediments of the same type. See Table 45 A for units.

Table 43 B. The effects of fresh crude oil at 50 ppt on various microbial processes in sediments amended with Cerophyl, chitin, or or starch after 1.5 years exposure.

	CONTROL				OILED			
Variable	Control	Cerophyl	Chitin	Starch	Control	Cerophyl	Chitin	Starch
CO_2 Evolut.	4.1	33.4	33.6	30.5	8.0	24.0	6.1	60.9
% Change					+95	-28	-82	+100
N_2 Fixat.	2.48	1.01	0.94	0.85	0.30	0.06	0.01	0.21
% Change					-88	-94	-99	-75
Methane	0.3	1.8	0.2	1.1	3.8	241	500	5.3
% Change					+346	+13364	+312400	+395
Glucose Upt.	104	160	93	97	74	197	174	147
% Change					-28	+23	+88	+52
% Respir.	17	28	28	28	39	48	49	65
% Change					+121	+72	+71	+131
Denitrif.	153	47	137	154	13	7	8	8
% Change					-92	-85	-94	-95
Phosphatase	0.38	0.44	0.38	0.37	0.22	0.38	0.33	0.39
% Change					-41	-14	-15	+6
Chitobiase			0.46				0.02	
							-48	
Redox potential	-70	-56	-97	-252	-430	-428	-415	-424
% Change					-514	-664	-328	-68

See Table 45 A for units.

Table 44. The effects of fresh oil at 50 ppt on various microbial processes in a sediment amended with Cerophyl, chitin, or starch after 12 mo. exposure. This sediment was collected in Sadie Cove.

	CONTROL				OILED			
Variable	Control	Chitin	Cerophyl	Starch	Control	Chitin	Cerophyl	Starch
CO_2 Evol.	46	37	68	44	23	137	97	92
% Change					-51	+270	+42	+112
N_2 Fixat.	3.2	1.4	2.4	1.3	0.1	0.07	0.5	0.3
% Change					-97	-95	-77	-80
Methane	6.4	4.5	22	6.3	4.0	128	107	33
% Change					-37	+2700	+390	+430
Glucose Upt.	442	306	120	276	74	68	128	41
% Change					-83	-78	+7	-85
Gls. % Resp.	18	17	17	18	39	47	61	34
% Change					+122	+169	+268	+91
Acetate Upt.	357	226	416	220	47	37	33	46
					-87	-83	-92	-79
Acet. % Resp.	24	22	24	22	30	59	44	30
% Change					+24	+169	+88	+41
Denitrif.	18	24	16	16	7	1	6	6
% Change					-61	-96	-62	-62
Phosphatase	0.23	0.23	0.25	0.14	0.16	0.17	0.14	0.18
% Change					-31	-24	-41	+30
Chitobiase		0.027				0.020		
% Change						-25		
Redox Potent.	-220	-242	-173	-196	-427	-422	-337	-316
% Change					+94	+74	+362	+61

% Change compares oiled and nonoiled samples of the same type.

See Table 45 A for units.

experiments in which there was only one sample for each treatment. The second set of observations was made in a subsequent set of experiments in which everything was triplicated (Tables 45 and 46). For this reason, we were able to assign levels of confidence only in the latter experiments.

When the nonaugmented sediments were analyzed, we observed the same type of changes in the crude oil treated sediments that we have described in the preceding sections; i.e., glucose uptake rates were depressed, respiration percentages increased (decreased microbial biosynthesis), denitrification and nitrogen fixation rates decreased, redox potentials decreased, methane concentrations increased, and phosphatase activity was depressed.

In all four studies, starch was added to some of the sediments analyzed. When we compared the oiled and nonoiled sediments, we found that the CO_2 evolution rates, the percent respiration and amylase activity values increased and that denitrification rates, phosphatase activity and redox potential values were reduced. In both of the pilot studies, the nitrogen fixation rates were depressed in the oiled sediments that had been augmented with starch. In the second two studies, (Tables 45 and 46), there was an increase in nitrogen fixation rates which was not statistically significant.

Chitin was another organic supplement which was utilized in all four studies. When similar measurements were conducted on these sediments, we found that the percent respiration increased and glucose uptake rates and the phosphatase and chitobiase activitive decreased. The nitrogen fixation rates in the pilot studies were depressed in the presence of crude oil and in the second set of experiments, the

Table 45. The effects of fresh crude oil at 50 ppt on various microbial processes in a sediment amended with starch, cellulose, chitin or seaweed after 8 mo. exposure. All measurements were made on triplicate subsamples.

		Glucose												Redox Potential	
AMENDMENT	TREATMENT	Uptake	% Resp.	CO_2	CH_4	N_2Fix	Denit.	Phos.	Amyl.	Alg.	Cell.	Lam.	Chit.	Surf.	Bot.
None	Control	105	18	11	0.15	2.5	258	0.53						-49	-94
	Oiled	38	36	10	3	0.4	76	0.30						-372	-412
	% Change	-49**	+100***	-11#	+1700*	-84**	-71**	-44**						-659***	-338***
Starch	Control	283	56	77	233	0.7	97	0.32	169					-293	-379
	Oiled	200	66	128	589	4.5	30	0.27	428					-399	-408
	% Change	-29#	+18*	+67#	+53**	+543	-69	-13*	+153***					-16#	-8**
Cellulose	Control	233	39	112	281	2.6	46	0.33			28			-261	-401
	Oiled	79	46	22	11	0.4	95	0.29			14			-365	-409
	% Change	-66**	+18#	-80**	-96**	-84#	+65**	-13#			-50**			-40#	-2#
Chitin	Control	91	35	64	59	0.3	101	0.46					0.062	-286	-397
	Oiled	58	46	19	1	0.4	48	0.31					0.026	-402	-425
	% Change	-36*	+31**	-71**	-98**	+40#	-52*	-34**					-58***	-41***	-7*
Seaweed	Control	163	27	35	11	0.9	164	0.45		30		22		-202	-304
	Oiled	93	53	15	69	0.05	25	0.27		57		14		-344	-436
	% Change	-43**	+96***	-56*	+554#	-94#	-85**	-40**		+90**		-36**		-70*	-43**

See Table 45 A for footnotes on Tables 45 and 46.

Table 45 A. Footnotes for Tables 45 and 46.

% Change compares oiled and nonoiled samples of the same type.

Variable	Units	Statistical Significance
Glucose uptake	ng x g^{-1} x h^{-1}	p less than
CO_2 = CO_2 production	nmole x g^{-1} x h^{-1}	* = 0.05
CH_4 = Methane evolution	pmolw x g^{-1} x h^{-1}	** = 0.01
N_2Fix = Nitrogen fixation	ng N x g^{-1} x g^{-1} x h^{-1}	*** = 0.001
Denit. = Denitrification rates	ng N x g^{-1} x h^{-1}	# = p >0.05
Phos. = Phosphatase activity	μmoles p-nitrophenol x g^{-1} x h^{-1}	
Amyl. = Amylase activity	μg reducing sugar x g^{-1} x h^{-1}	
Alg. = Alginase activity	" " " " "	
Cell. = Cellulase activity	" " " " "	
Lam. = Laminarinase activity	" " " " "	
Chit. = Chitobiaseactivity	μmoles p-nitrophenol x g^{-1} x h^{-1}	
Redox potential	mv	
Surf. = sampled at sediment surface		
Bot. = " " ca. 8 cm depth		

Table 46. The effects of fresh crude oil at 50 ppt on various microbial processes in a sediment amended with starch, cellulose, chitin or gelatin after 6 mo. exposure. All measurements were made on triplicate subsamples.

AMENDMENT	TREATMENT	Glucose										Redox Potential	
		Uptake	% Resp.	CO_2	CH_4	N_2Fix	Denit.	Phos.	Amyl.	Cell.	Chit.	Surf.	Bot.
None	Control	125	18	16	0.7	1.3	205	0.27				-1	-130
	Oiled	64	41	24	1.0	0.2	106	0.17				-314	-418
	% Change	-49**	+128***	+50*	+43#	-85***	-48**	-35***				-31300***	-222***
Starch	Control	347	59	150	181	0.05	5	0.33	168			-289	-404
	Oiled	349	72	421	171	6.6	5	0.27	310			-348	-387
	% Change	+1#	+22**	+181**	-6#	+1310#	0#	-18*	+85**			-20*	+4#
Cellulose	Control	122	37	126	60	0.27	36	0.37		31		-372	-415
	Oiled	60	52	17	0.1	0.12	81	0.22		13		-368	-372
	% Change	-51#	+41**	-87*	-99**	-55#	+125**	-39**		-58**		+1#	+10#
Chitin	Control	66	38	43	46	0.12	12	0.38			0.68	-296	-421
	Oiled	71	52	46	1.4	0.40	106	0.42			0.15	-297	-366
	% Change	+7#	+37**	+5#	-97*	+233#	+783***	+10#			-78***	0#	+13**
Gelatin	Control	68	31	34	98	0.30	70	0.32				-344	-413
	Oiled	25	61	23	348	0.09	7	0.18				-395	-436
	% Change	-63***	+97***	-29#	+155#	-70#	-90*	-45***				-15#	-6#

nitrogen fixation rates increased but this change was not statistically significant.

In the two pilot studies, a third substrate was tested; Cerophyl. This is a commercial preparation of dried grass. In the Sadie Cove sediment and in the first observations made in the Kasitsna Bay sediment, the CO_2 production rates were higher in the oiled sediments (Tables 43 and 44). In the Kasitsna Bay sediment after 18 month exposure, the nitrogen fixation rate was depressed in the oiled sediment. We also observed a reduction in denitrification rates, acetate uptake rates, and the phosphatase and cellulase activities. Increases were observed in glucose uptake rates, percent respiration with both acetate and glucose, and methane concentrations.

In the second two studies, cellulose was used in place of Cerophyl. In these sediments, the glucose uptake, nitrogen fixation rates, methane concentrations, and the cellulase activity rates were lower in the oiled sediments. Increases were observed in the percent respiration, and in the denitrification rates. Two other substrates were also tested in the second set of observations; dried seaweed and gelatin. In the sediments supplemented with seaweed, we observed an increase in percent respiration, methane concentration, and alginase activity and a reduction in the glucose uptake rate, CO_2 production, nitrogen fixation and denitrification rates and phosphatase and laminarinase activity in the oiled sediments. In the sediments supplemented with gelatin, we observed in increase in percent respiration, and methane concentration. We also observed a decrease in the glucose uptake rate, phosphatase activity and the rates of nitrogen fixation and denitrification.

During the second set of observations, we also measured the redox potentials at both the surface and bottom of the undisturbed sediments. The results of those measurements are shown in Table 45 and 46. In all cases, the augmented sediments had much lower redox potentials than did the nontreated sediments.

There were also other consistent changes which took place in the sediment augmented with organic nutrients when compared with the control values. As shown on Tables 45 and 46, the percent respiration increased in all sediments that were augmented with organic carbon. Both methane concentrations and CO_2 evolution rates were consistently much greater and nitrogen fixation and denitrification rates were generally lower in the augmented samples.

C. Seasonal observations of microbial function in the sediments and waters near Kasitsna Bay.

1. Water column observations including relative microbial activity, primary productivity and inorganic nutrient data.

During the course of our work at Kasitsna Bay, we measured relative microbial activity in water samples collected at the locations shown in Fig. 37. These determinations were made using labeled glucose, glutamic acid, acetate and glycollate (Table 47 A and 47 B). During the winter months, the microbial activities were very low; in the spring or early summer, there was an increase with maximum rates observed in April in 1979 and August in 1980 as measured using glucose or glutamic acid. These were also when the uptake ratios as measured using glucose/glutamic acid uptake rates were the highest. Most of these observations were made at the same 15 stations located near Kasitsna Bay. Five of these stations were routinely sampled for primary productivity measurements.

Table 47 A. Primary productivity, glycollate uptake and acetate uptake in water samples from field stations at Kasitsna Bay.

		1-80	4-4-80	4-13-80	4-21-80	4-28-80	6-80	7-80	8-80
	$\bar{Y}$	0.3	0.9	1.4	4.3	3.8	9.2	4.6	7.2
Primary	SD	0.2	0.6	0.7	4.6	2.4	5.4	3.4	4.6
Prod.	Range	.2-.6	0.4-1.8	0.6-2.4	1.2-2.4	1.5-7.3	0.3-13.9	0.3-8.1	3.7-15.2
	$\bar{Y}$		11	26	18	19	21	83	92
Glycollate	SD		6	9	7	7	11	68	86
Uptake	Range		4-23	17-40	11-28	12-29	4-32	37-204	20-191
	N		12	5	5	5	5	5	5
	$\bar{Y}$			4	4	4	15	73	30
Acetate	SD			1	1	2	7	62	15
Uptake	Range			3.0-4.6	2.6-4.5	1.8-7.0	6.4-22	33-183	18-54
	N			5	5	5	5	5	5

	Units
Primary productivity	μg C/meter^3
Glycollate uptake	ng x l^{-1} x h^{-1}
Acetate uptake	ng x l^{-1} x h^{-1}

Table 47 B. Glucose and Glutamate uptake in water samples from field stations at Kasitsna Bay.

		2-79	4-79	7-79	8-79	11-79	1-80	4-4-80	4-13	4-21	4-28	6-80	7-80	8-80
	$\bar{Y}$	0.7	135	86	29	5	2	2	11	9	10	80	193	238
Glucose	SD	0.3	131	57	20	3	1	1	11	9	9	68	149	180
	Range	04.-1.2	11-366	22-181	6-65	1-10	1.2-3.6	8-4.0	0.6-28.4	3.0-25.5	1.4-21.8	10.29-232.89	69.28-447.82	95.9-528.7
	N	7	6	15	15	17	10	12	5	5	5	13	5	5
	$\bar{Y}$	7	134	101	-	12	6	7	22	23	18	86	217	233
Glutamate	SD	6	115	74	-	5	3	4	20	9	17	41	223	149
	Range	2-18	32-170	21-245	-	3-23	3-23	3-14	1.7-16.7	2.6-49.5	8.6-32.0	4.4-42	93.57-613.88	76.6-397.1
	N	7	6	15	-	15	10	12	5	5	5	13	5	5
Ratio means Gluc./Glut.		.10	1.01	.85		.42	.33	.29	.50	.39	.55	.93	.89	1.02

	Units
Glucose uptake	ng x l^{-1} x h^{-1}
Glutamate uptake	ng x l^{-1} x h^{-1}

The primary productivity rates observed during the 1980 field season are also listed in Table 47 A. During this season, the lowest rates were observed in January and the highest were in June. Linear regression analysis was performed on the entire data set which included primary productivity rates and uptake rates for glucose, glutamate, acetate, and glycollate (Table 48). All the uptake variables were very highly intercorrelated ($p<0.001$). Primary productivity was significantly correlated ($p<0.05$) with glutamate uptake; highly correlated ($p<0.01$) with glucose uptake; and very highly correlated with the ratio of glucose/glutamate uptakes. The graphic presentation of seasonal glucose uptake and primary productivity rates indicates that the increase in primary productivity rates during the 1980 season preceeded the increase in glucose uptake rates in the spring (Fig. 61).

During the 1979 field season, measurements of inorganic nutrient concentrations were conducted (Table 49). In general, the concentrations observed were highest in the winter and lowest in the summer.

2. Observations made on the sediments.

a. Relative microbial activity and nitrogen fixation rates.

During the course of our Kasitsna Bay work, we measured relative microbial activity using glucose and glutamic acid (Table 50). The lowest rates were observed in February and April 1979 and the highest rates were observed in August 1979. This was true when relative microbial activities were measured with either glucose or glutamic acid. As was the case in the water samples, the ranges

Table 48. Linear correlation coefficients (r-value) for surface water microbial activities at Kasitsna Bay field stations sampled from Feb. 1979 thru Aug. 1980.

	Primary Productivity	Glucose uptake	Glutamate uptake	Acetate uptake	Glucollate uptake
Glucose uptake	0.40 ** (38)				
Glutamate uptake	0.34 * (38)	0.89 *** (113)			
Acetate uptake	0.06 (28)	0.71 (28)	0.87 (28)		
Glycollate uptake	0.27 (33)	0.86 *** (39)	0.95 *** (39)	0.76 *** (39)	
Glucose/Glutamate	0.58 *** (38)	0.54 ** (113)	0.26 ** (113)	0.19 * (28)	0.26 ** (39)

Significance levels:		
	*	p ≤0.05
	**	p ≤0.01
	***	p ≤0.001

( ) - degrees of freedom

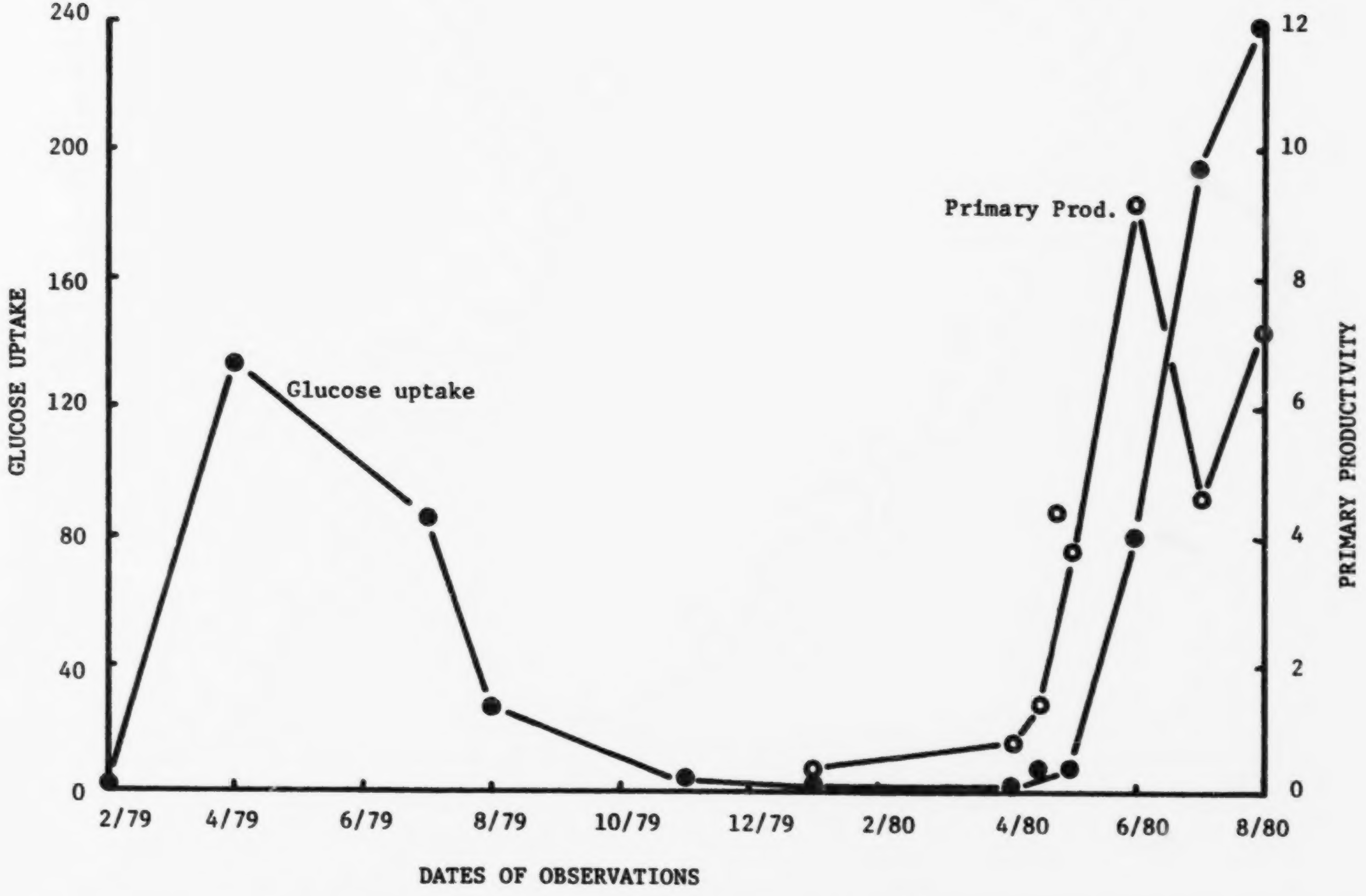

Figure 61. Seasonal changes in glucose uptake (ng x l^{-1} x h^{-1}) and primary productivity (µg carbon x m^2 x h^{-1}) in water samples collected near Kasitsna Bay.

Table 49. Nutrient analysis of surface waters from Kasitsna Bay field stations sampled from Feb. 1979 thur Jul. 1979.

		Feb. 1979	Apr. 1979	Jul. 1979
NH_4^+ (µm)	$\bar{Y}$	.87	.94	1.2
	sd	.11	.25	.84
	range	.74-1.0	.67-1.4	.14-3.0
	n	7	6	28
NO_3^- (µm)	$\bar{Y}$	13.5	2.0	1.3
	sd	1.1	1.7	1.4
	range	12-15	0-4.3	0-4.5
	n	7	6	28
NO_2^- (µm)	$\bar{Y}$	.09	.04	.03
	sd	.01	.05	.04
	range	07-.1	0-.15	0-.14
	n	7	6	28
PO_4^{-3}(µm)	$\bar{Y}$	1.4	.63	.61
	sd	.07	.15	.33
	range	1.3-1.5	.4-.82	.2-1.4
	n	7	6	28

Table 50. Uptakes of Kasitsna Bay sediments.

		DATE						
		2-79	4-79	8-79	11-79	1-80	4-80	6-80
	$\bar{Y}$	25	27	140	57	46	53	50
Glucose		29	22	94	57	30	41	44
Uptake	Range	3-	5-	20-	4-	11-	10-	9-
		110	73	360	180	120	140	150
	N	17	10	18	15	10	12	11
	$\bar{Y}$	340	320	1070	320	380	380	470
Glutamate	SD	425	250	970	330	280	200	260
Uptake	Range	32-	35-	180-	41-	110-	130-	120-
		1860-	940	3600	1260	1110	780	960
	N	17	10	18	15	10	12	10

	Units
Glucose uptake	ng x g. dry wt.$^{-1}$ x h^{-1}
Glutamate uptake	" " " " "

in values were greatest when glucose was used as the test substrate. The ranges observed in the sediments were much less than those observed in the water samples during the period of our study.

In the region near Kasitsna Bay, there were two major sources of terrestrial carbon input at the head of each of the major bays. The pattern of relative microbial activity in the sediment samples of the area reflects the input of terrestrial carbon. When we compare the relative microbial activity as measured with glucose in the sediments taken near the head of these bays with sediments taken from other locations, the relative microbial activity near the head of the bays was significantly higher (Table 51). This was true during all of the field seasons.

A different pattern was obsrved when the sediments of Kachemak Bay were analyzed for relative microbial activity (Fig. 62). In Kachemak Bay, the highest values were observed seawater of Homer spit and the lowest values were observed near the mouth of the Fox River. The same pattern was observed when glutamic acid was used to measure relative microbial activity.

b. Nitrogen fixation rates, total adenylate concentration, and inorganic nutrient concentrations.

The mean nitrogen fixation values observed in August 1979 were the lowest and those observed in July 1980 were the highest measured during the course of our field station studies (Table 52). The differences observed between the August 1979 and the high values in October 1979 and July 1980 were statistically significant. The only strong geographical patterns of nitrogen fixation that were

Table 51. Differences in glucose uptake rates between sediments from sediments collected from Kasitsna Bay field stations near the heads of the bays compared with sediments collected more seaward.

Date	Bay	Seaward	Diff.	%	P<
4-79	42	14	28	-67	0.05
7-79	224	99	125	-56	0.01
10-79	101	27	74	-73	0.05
4-80	96	31	65	-68	0.01
6-80	114	27	87	-76	0.001

Glucose uptake ng x g. dry wt.$^{-1}$ x h^{-1}

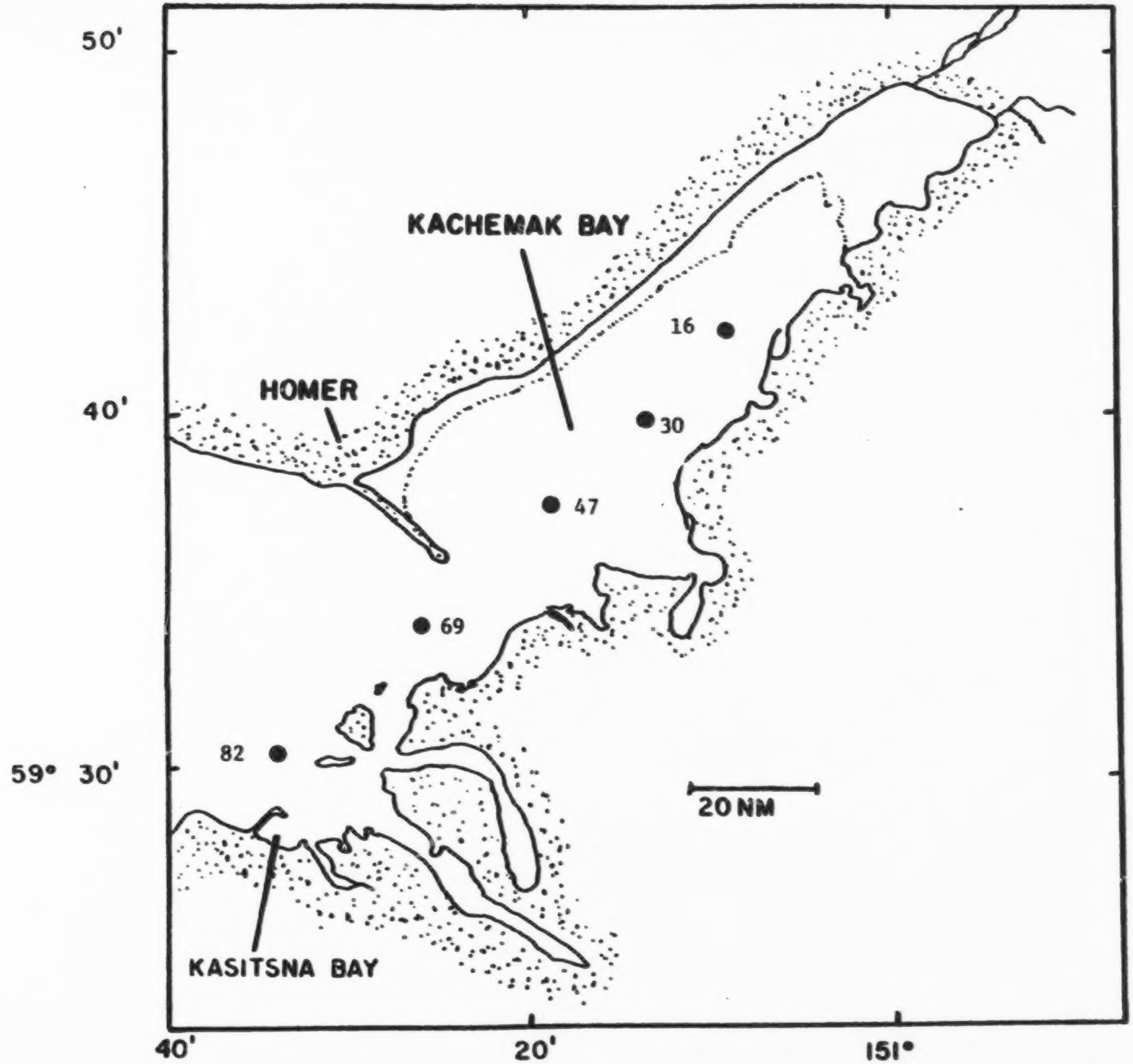
50'
40'
59° 30'
40'
20'
151°
KACHEMAK BAY
HOMER
KASITSNA BAY
16
30
47
69
82
20 NM

Figure 62. Glucose uptake rates in the sediments of Kachemak Bay reported as ng x g^{-1} x h^{-1}.

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Table 52. Summary of nitrogen fixation rates, total adenylate concentrations and energy charge estimates in Kasitsna Bay sediments.

A. Nitrogenase activity[1]

Date	$\bar{Y}$	SD	Range	n
Feb. 1979	0.7	0.6	0-2.3	11
Apr. 1979	0.9	0.8	-2.1	11
July 1979	2.4	1.4	0.9-4.2	4
Aug. 1979	0.4	0.4	0-1.0	14
Oct. 1979	1.7	1.9	0-7.5	14
Jan. 1980	0.9	0.5	0.3-1.8	8
Apr. 1980	0.9	0.5	0.3-1.8	9
July 1980	1.3	0.8	0.2-3.1	11

B. Adenylate determinations

	Total Adenylates[2]			Energy Charge			
Date	$\bar{Y}$	SD	Range	$\bar{Y}$	SD	Range	n
Feb. 1979	4.4	2.3	0.1-7.3	0.31	0.16	0.18-57	9
Apr. 1979	6.9	1.9	4.2-8.5	0.29	0.04	0.24-0.35	4
Jul. 1979	7.7	3.1	3.3-12.9	0.26	0.08	0.12-0.38	14

[1] ngN$_2$/g dry wt/hr.

[2] nmoles/g.

observed occurred in the sediment samples collected along the middle of Kachemak Bay (Fig. 63).

The concentration of total adenylates increased from a low of 4.4 nM/g dry wt in February 1979 to a high of 7.7 nM/g dry wt in July 1979 (Table 52). The calculated energy charge values decreased from a high of 0.31 in February to a low of 0.26 in July. These differences were not statistically significant.

Analyses of inorganic nitrogen species and phosphate concentrations in pore water and total carbon, nitrogen and phosphorus concentrations in sediment samples were also conducted for all field stations (Table 53). The results of the total sediment chemistry showed very little seasonal variation; however, seasonal changes were observed in the pore water chemistry. Both NH_4^+ and PO_4^{-3} concentrations were highest during the month of April 1979 and lowest during the month of July 1979.

c. Enzyme activities observed in sediments

During the field station work, we routinely measured the activities of the following enzymes: amylase, cellulase, phosphatase, and arylsulfatase (Table 54). In general, the activity levels observed were constant throughout the year. The most significant geographical trend that was noted was in the arylsulfatase/phosphatase ratios. In both the Kasitsna Bay stations (Fig. 64) and the Kachemak Bay stations (Fig. 65), these ratios generally increased in sediment samples collected increasingly further from major fresh water sources.

A linear regression analysis to determine enzyme activity correlations with glucose and glutamic acid uptake and nitrogen fixation rates was performed for Kasitsna Bay field stations (Table 55).

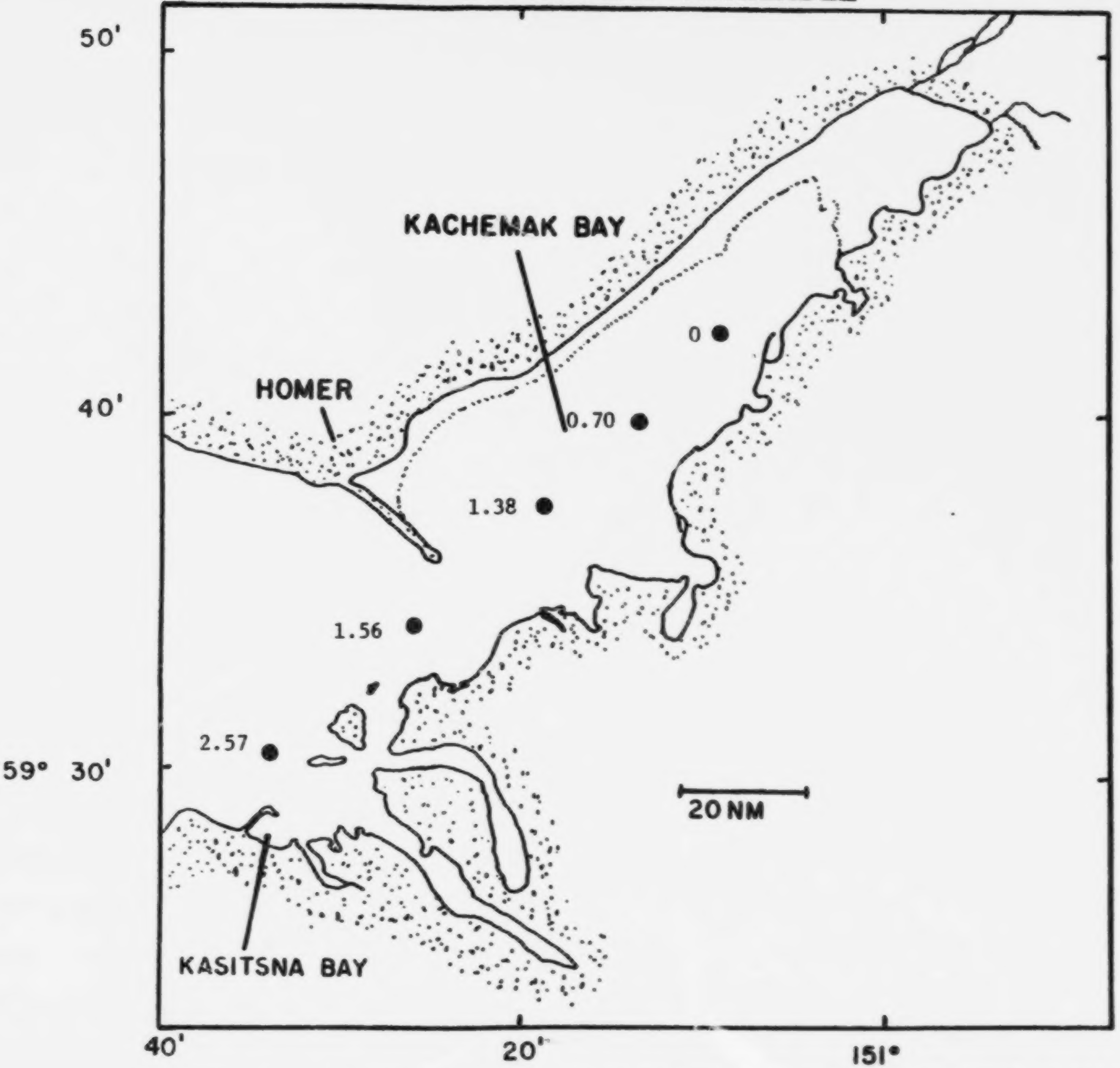
50'
40'
59° 30'
40'
20'
151°
KACHEMAK BAY
HOMER
KASITSNA BAY
0
0.70
1.38
1.56
2.57
20 NM

Figure 63. Nitrogen fixation rates in Kachemak bay sediments reported as ng x g^{-1} x h^{-1}.

Table 53. Nutrient analysis of sediments from Kasitsna Bay field stations sampled from Feb. 1979 through Jan. 1980.

		Interstitial Water				Total Sediment				
Date		NO_3 + NO_2 um	NH_4 um	NO_2 um	PO_4 um	P ppt	C ppt	N ppt	$\bar{C}/\bar{N}$	$\bar{C}/\bar{P}$
2-79	$\bar{Y}$	1.7	210	1.5	39					
	S.D.	1.7	130	1.2	38					
	Range	0-	72-	0-	11-					
		5.6	300	3.9	74					
	n	16	16	16	16					
4-79	$\bar{Y}$	1.6	680	1.9	110	.9	25	2.0	13	30
	S.D.	1.6	852	1.2	100	.1	10	.7		
	Range	0-	68-	.4-	16-	.7-	12-	.6-		
		4.4	2700	4.0	360	1.0	41	3.3		
	n	11	11	11		11	11	11		
7-79	$\bar{Y}$	1.1	100	1.2	15	.8	24	1.9	12	29
	S.D.	.8	58	1.7	13	.2	9.5	.7		
	Range	0-	48-	0-	5.8	.5-	11-	1.1-		
		2	220	5	41	1.2	41	3.5		
	n	7	7	7	7	20	20	20		
11-79	$\bar{Y}$					.7	23	2.0	12	33
	S.D.					.1	9.1	1.2		
	Range					.5-	8.7-	.6-		
						.8	38	4.7		
	n					14	14	14		
1-80	$\bar{Y}$					.7	27	2.5	11	36
	S.D.					.1	11	.5		
	Range					.6-	14-	1.7-		
						.9	45	3.1		
	n					5	5	5		

Table 54. Summary of sediment enzyme data for Kasitsna Bay field stations.

A. Amylase activity (μg glucose released x g dry wt^{-1} x h^{-1})

Date	$\bar{Y}$	sd	Range	n
Feb. 1979	31.5	14.8	10.8 - 62.2	12
Apr. 1979	28.1	22.4	10.8 - 82.6	11
Jul. 1979	25.8	12.2	10.1 - 45.4	13
Oct. 1979	31.6	27	10.6 - 99.7	14
Jan. 1980	18.7	13	6.3 - 51	10
Apr. 1980	29.5	17.9	14.3 - 75.4	12
Jun 1980	27.9	18.1	10.4 - 72.5	11

B. Cellulase activity (μg glucose released x g dry wt^{-1} x h^{-1})

Date	$\bar{Y}$	sd	Range	n
Feb. 1979	15.9	8.4	4.3 - 31.4	12
Apr. 1979	14.3	7.8	6.5 - 35.3	11
Jul. 1979	17.5	9.9	8.1 - 37.6	13
Oct. 1979	11.8	12	1.7 - 48.7	14
Jan. 1980	10.3	4.7	3.7 - 20	10
Apr. 1980	14.2	7.1	7.9 - 33.4	12
Jun. 1980	20.5	11.7	7.5 - 42.8	11

C. Phosphatase activity (μM p-nitrophenol released x g dry wt^{-1} x h^{-1})

Date	$\bar{Y}$	sd	Range	n
Feb. 1979	0.27	0.23	0.03 - 0.81	12
Apr. 1979	0.22	0.09	0.13 - 0.41	11
Jul. 1979	0.31	0.11	0.14 - 0.48	14
Oct. 1979	0.27	0.14	0.15 - 0.62	14
Jan. 1980	0.28	0.13	0.14 - 0.50	10
Apr. 1980	0.23	0.09	0.12 - 0.44	12
Jun 1980	0.34	0.14	0.19 - 0.70	11

(continued on following page)

Table 54 (continued)

D. Arylsulfatase activity (μM p-nitrophenol released x g dry wt^{-1} x h^{-1})

Date	$\bar{Y}$	sd	Range	n
Feb. 1979	0.50	0.43	0.01 - 1.41	12
Apr. 1979	0.37	0.16	0.08 - 0.62	11
Jul. 1979	0.60	0.24	0.30 - 1.12	14
Oct. 1979	0.48	0.26	0.09 - 1.04	14
Jan. 1980	0.63	0.26	0.21 - 1.13	10
Apr. 1980	0.41	0.21	0.14 - 0.85	12
Jun. 1980	0.56	0.30	0.16 - 1.19	11

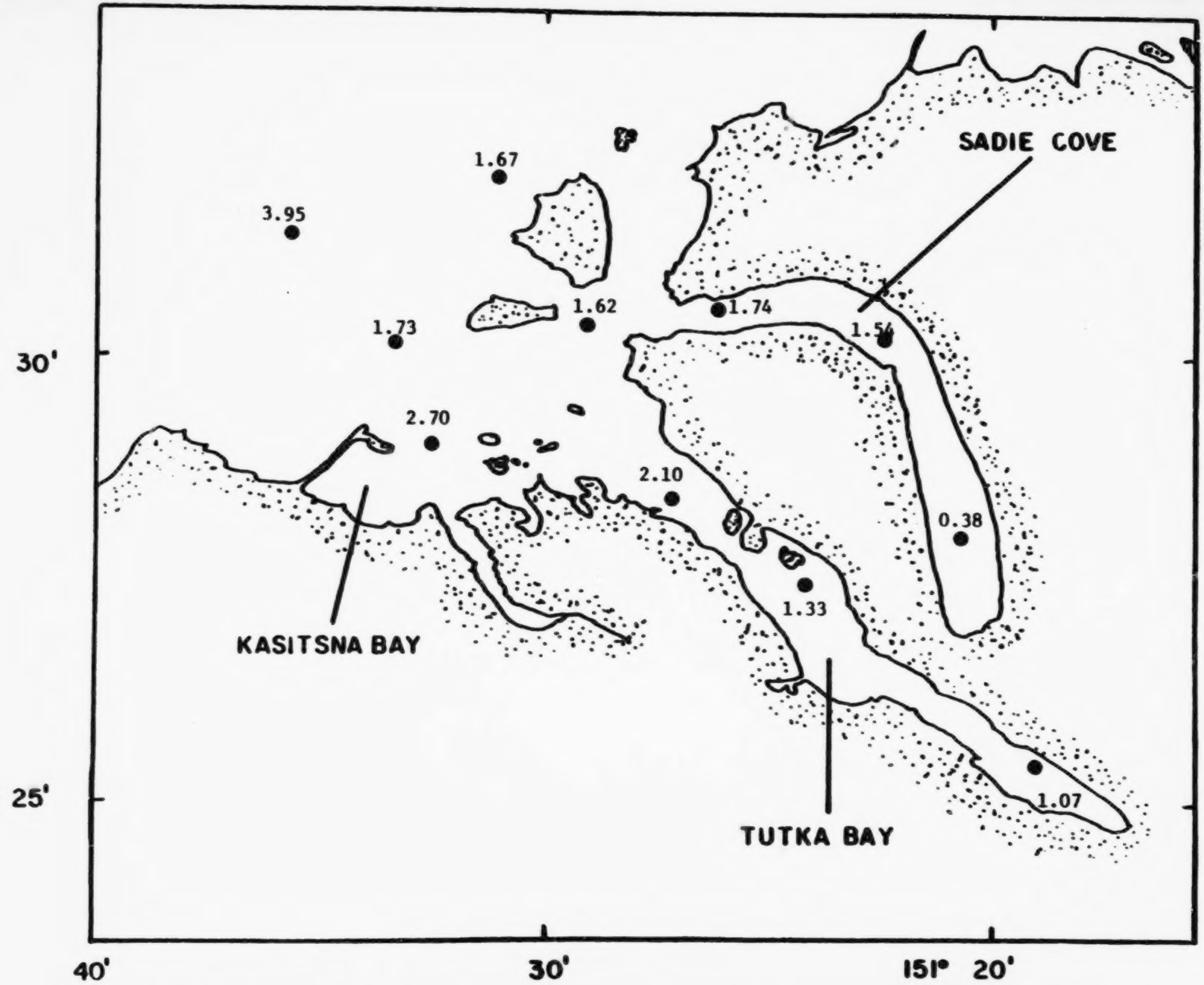
SADIE COVE
1.67
3.95
1.62
1.74
1.73
1.56
30'
2.70
2.10
0.38
1.33
KASITSNA BAY
1.07
59° 25'
TUTKA BAY
40'
30'
151° 20'

Figure 64. The ratios of arlysulfatase/phosphatase activity in sediments near Kasitsna Bay.

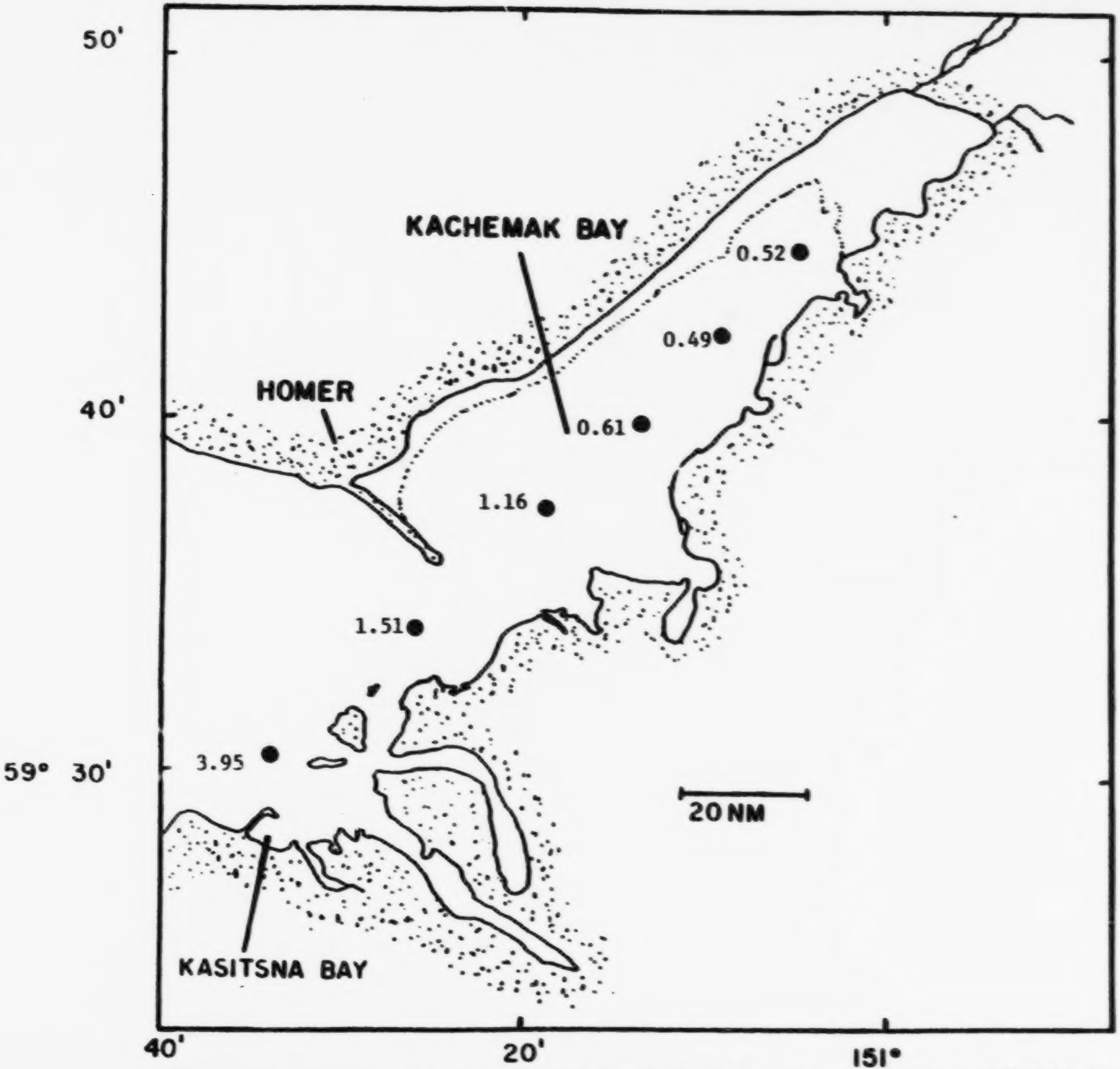
50'
40'
59° 30'
40'
20'
151°
KACHEMAK BAY
HOMER
KASITSNA BAY
0.52
0.49
0.61
1.16
1.51
3.95
20 NM

Figure 65. The ratios of arylsulfatase/ phosphatase activity in sediments in Kachemak Bay.

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Table 55. Linear correlation coefficients (r-value) for sediment microbial activities at 83 Kasitsna Bay field stations sampled from Feb. 1979 through Aug. 1980.

	Amylase	Cellulase	Phosphatase	Arylsulfatase	Glucose uptake	Glutamate uptake
Cellulase[1]	0.76 ***					
Phosphatase[1]	0.16	0.23 *				
Arylsulfatase[1]	0.14	0.24 *	0.81 ***			
Glucose uptake[2]	0.25 *	0.31 **	0.29 **	0.25 *		
Glutamate uptake[2]	0.03	0.01	0.27 *	0.16	0.74 ***	
N_2 fixation[2]	0.30 **	0.34 **	0.25 *	0.21	0.19	0.05

Significance levels: * $p \leq 0.05$
** $p \leq 0.01$
*** $p \leq 0.001$

[1] - enzyme inter-correlations for 81 degrees of freedom

[2] - other correlations for 79 degrees of freedom

The highest correlations observed were between amylase and cellulase activity, phosphatase and arylsulfatase activity, and glucose and glutamic acid uptake rates.

During the April 1980 sampling period, the 12 field sediments sampled were tested for redox potential, CO_2 evoluation and CH_4 content in addition to the standard assays. A linear regression analysis on this data set (Table 56) showed highly significant correlations between both CO_2 evolution and CH_4 content and the independent variables; amylase, cellulase, and glucose/glutamate uptake ratio. Redox potential was correlated only with CO_2 evoluation.

Table 56. Linear correlation coefficients (r-value) for sediment CO_2, CH_4 and Redox values at 12 Kasitsna Bay field stations sampled in Apr. 1980.

	CO_2	CH_4	Redox
Amylase	0.89 ***	0.81 **	0.47
Cellulase	0.82 **	0.86 ***	0.54
Phosphatase	0.58 *	0.29	0.14
Arylsulfatase	0.27	0.11	0.50
Glucose uptake	0.63 *	0.42	0.48
Glutamate uptake	0.21	0.16	0.30
Glucose/Glutamate	0.82 ***	0.91 ***	0.47
CO_2 Evolution		0.78 **	0.59 *
CH_4 Concentration			.33

Significance levels:
* $p \leq 0.05$
** $p \leq 0.01$
*** $p \leq 0.001$

V. Discussion

A. Short-term effects of crude oil and/or Corexit 9527.

1. Crude oil effects

a. There have been a number of studies (Alexander and Schwarz, 1980; Atlas et al., 1976; Calder and Lader, 1976; Griffin and Calder, 1977; Hodson et al., 1977; and Walker and Colwell, 1975) that have addressed the question: what effects do crude oil, refined petroleum products, or pure hydrocarbons have on marine microbial populations? Several of these studies were concerned with the effects of crude oil on microbial growth rates. For the most part, these studies have shown that crude oil has very little effect on growth; however, reduced growth rates have been shown in populations exposed to refined petroleum products (Walker and Colwell, 1977) or aromatic hydrocarbons (Calder and Lader, 1976; Griffin and Calder, 1977). In a study of the effects of crude oil on cell numbers in seawater taken from Yaquina Bay, Oregon, no significant changes in plate counts were observed in water samples exposed to Prudhoe Bay crude oil for periods of up to 40 days (Griffiths, unpublished data). Atlas et al. (1976) reported that Prudhoe Bay water exposed to crude oil showed an increase in cell number relative to the control after 42 days. Although no significant change in species diversity was observed in this experiment, a more recent study by Atlas (personal communication) using numerical taxonomical techniques has shown shifts in the composition of bacterial populations in Arctic marine waters exposed to Prudhoe crude oil for several months. These data suggest that although populations of marine heterotropic bacteria may not decrease when exposed to crude oil, the crude oil may act as an environmental stress which could affect species composition and thus microbial function.

Hodson et al. (1977) reported that crude oil-aqueous solutions inhibited the uptake and mineralization of ^{14}C labeled glucose by pelagic microorganisms. These observations were made in seawater samples contained in large plastic bags during the course of the CEPEX project (Menzel and Case, 1977). In our study, a large number of pelagic and benthic microbial populations collected from a wide variety of sources were exposed to crude oil. The uptake and mineralization of labeled glucose was monitored to determine if altered function could be detected.

b. Both this study and that reported by Hodson et al. (1977) indicate that there is a significant reduction in the heterotrophic uptake and mineralization rates in pelagic microbial populations when they are first exposed to crude oil and/or refined oils. This effect was observed when either glucose or glutamic acid was used as the test substrate indicating that this phenomenon is not restricted to glucose (the substrate used by Hodson). The phenomenon, caused by a number of crude oil type products, affects a large cross section of natural marine microbial populations; although pelagic populations appear to be affected to a greater extent than benthic populations. In a recent study by Alexander and Schwarz (1980), the short-term effects of two crude oils on glucose uptake in pelagic and benthic microbial populations was observed. They observed no consistent depression of glucose uptake in the presence of crude oil. It is quite possible that their samples, taken from Galveston Bay and the Louisiana coasts had been exposed to chronic petroleum perturbation prior to collection.

c. There was a wide variation found in the percent reduction observed from one location to another. The geographic location of the samples showing these variations indicate that the degree to which a population is affected may be related to the degree of prior exposure to crude oil or petroleum products.

d. Even though there is an initial reduction of relative microbial activity in natural marine microbial populations exposed to crude oil, there is evidence that suggests pelagic microorganisms may adjust to the presence of petroleum. During one cruise in the Cook Inlet, there were differences noted in the extent to which natural populations were affected by the presence of crude oil in acute challenge experiments (Fig. 41). The patterns that emerged suggest that in regions where there should have been chronic input of petroleum hydrocarbons, the effects of crude oil were less than in the regions where this would not have been the case. For example, in the samples that were collected along the east-west transect from Kamishak Bay to Kachimak Bay, there was a reduced effect in the samples collected near the middle of Cook Inlet. One of the samples collected in this region showed stimulated glucose uptake in the oil-treated samples. The central samples were located in a region of little net water flow which is also in the center of the shipping line. It is assumed that petroleum products are constantly being introduced into these waters as the result of shipping.

Additional support for this hypothesis comes from the comparison of data collected to the north near Kalgin Island with the observations made to the south near Homer, AK. The water samples that were collected at the station near Homer showed a greater reduction in relative microbial activity when exposed to crude oil than did the samples collected in the two northern stations. The water mass at the sample site near Homer consists primarily of open ocean water which should have had little prior exposure to petroleum products. The northern sample sites are located near oil wells in the Inlet and also near an oil refinery with associated shipping facilities. Roubal and Atlas (1978) reported high concentrations of hydrocarbon-utilizing bacteria in the water samples collected at the two northern sample sites. There have been numerous reports in the literature which link elevated concentrations of hydrocarbon-utilizing bacteria with petroleum contamination in the marine environment (Horowitz and Atlas, 1977 and Mulkins-Phillips and Stewart, 1974).

During the same study, we observed that crude oil had very little effect on a water sample collected in Oil Bay where a natural oil seep has been documented. In an adjacent bay where a similar seep has not been observed, crude oil had a much greater effect on both glucose and glutamic acid uptake rates.

All of the observations listed above present only a presumptive correlation between prior exposure to petroleum and reduced sensitivity to the adverse acute effect of crude oil on relative microbial activity. More direct evidence has also been obtained from the observations discussed below.

e. In all of the above mentioned studies, the exposure time used was relatively short (normally under 12 h). When longer exposures were used (up to 9.5 days), the pelagic microorganisms appeared to adjust to the presence of crude oil (Figs. 39 and 40). In both studies, the level of relative microbial activity in the oiled water samples was lower than in nontreated samples at the beginning of the experiment. As the incubation time increased, the microbial activity in the oiled samples became greater than the controls. It should be pointed out that this phenomenon was only observed in water samples. We have demonstrated that in sediments which have been exposed to crude oil for extended periods of time, the relative mirobial activity in oiled sediments remains lower than the controls for exposure period of up to 1.5 years. It should also be said that the results of these experiments may not reflect what happens in the open ocean. It has long been recognized that the microbial characteristics of a seawater sample are greatly altered by confining it in a reaction vessel. This is the so called "surface" or "bottle" effect which causes the number of organisms present to multiply (within a few days) to form a population which could be 100 X the original concentration. This change in cell numbers is expressed as a significant increase in relative microbial activity after ca. 2 days incubation in both of these experiments.

The apparent adjustment of the pelagic microorganisms to the presence of crude oil was measured by only one type of function in this experiment; i.e., the uptake and utilization of a soluble organic substrate. This does not necessarily imply that there have been no shifts in the composition of the microbial population or that other microbial functions have not been altered.

f. Our studies have also shown that crude oil does not affect nitrogen fixation under short-term exposure. This is essentially the same conclusion reached by Knowles and Wishart (1977) in a similar study made on samples collected in and near the Beaufort Sea. The results of the short-term exposure experiments are in marked contrast to our findings of long-term crude oil effects on nitrogen fixation rates. The results of these studies are presented later in this section.

2. Corexit effects.

a. There has been growing evidence that dispersants may have adverse effects on a wide range of aquatic organisms. Hagstrom and Lonning (1977) showed that the dispersant (Corexit 9527) interfered with fertilization and development of a sea urchin with some adverse effects observed at concentrations as low as 0.0003 ppm. Hsiao et al. (1978) showed that crude oil-Corexit mixtures were inhibitory to primary production in Arctic marine phytoplankton. There has been very little information published concerning the impact of dispersants on microbial function. Gunkel (1968) states that 90 percent of marine bacteria were killed in the presence of 10 ppm "emulsifier". Mulkins-Phillips and Stewart (1974) showed that two of four dispersants studied slowed initial growth rates. This increased lag phase was more pronounced at an incubation temperature of 10 C than at 25 C.

b. Our Corexit acute effects studies have shown that this dispersant reduces the level of microbial activity in both water and sediment samples. In one set of observations, the concentration of Corexit required to reduce glucose uptake rates by 50% was 12 ppm. The reduction in substrate uptake rates is probably due to an induced stress on the microbial population. In the Mulkins-Philips and Stewart study (1974), it was shown that even though some dispersants caused an increase in the lag phase of growth, growth was not inhibited. When natural populations were exposed to dispersants and crude oil, however, there were qualitative shifts in the microbial population. In one case, the resulting population consisted of 100% pseudomonads. This was one of the experiments conducted during the CEPEX project described by Menzel and Case (1977) which consisted of observing the results of adding crude oil and Corexit 9527 to one of the seawater enclosure systems. At the end of the experiment, the microbial population was essentially a pure culture (G. G. Gessey, personal communication). It is quite possible that what we are observing in this study is the initial phase of a selection process which is acting to reduce the diversity of the population. The changes in respiration percentages that we observed in treated samples may reflect changes in the composition of the natural microbial populations.

c. As was the case in the crude oil effects experiments, the above-mentioned Corexit acute experiments were conducted with exposure times of 12 h or less. In order to determine if there was an apparent adjustment of pelagic microorganisms to the presence of Corexit, we conducted the same type of experiment with this pollutant as that used with crude oil (Figs. 44 and 45). We found that during the first 3 to 4 days exposure, the relative microbial activity was depressed in the treated water samples; however, as the incubation time increased, the relative microbial activity in the treated samples equalled and eventually surpassed that observed in the controls. Extremely high relative microbial activity levels were observed when both Corexit and nonlabeled glutamic acid were added to the water sample. This indicates to us that Corexit does not interfere with the growth of those organisms present which are capable of utilizing glucose (the labeled substrate used to determine relative microbial activity) and those capable of utilizing glutamic acid (the nonlabeled substrate).

Although these experiments are subject to the same potential problems of interpretation as the crude oil exposure experiments, there is stronger evidence that in fact, the organisms have adjusted to the presence of Corexit (at least as far as glucose uptake is concerned). Near the termination of both of these experiments, we added another 50 ppm of Corexit to the reaction mixture. Those samples which had previously been exposed to Corexit showed no additional reduction in relative microbial activity, whereas the controls (both with and without added NH_4^+) showed dramatic reductions (65-85%) in glucose uptake rates. One curious aspect of these experiments was that those samples to which nonlabeled glutamic acid was added showed no reduction in glucose uptake rates when exposed to the Corexit. These data suggest that actively growing cells which are capable of utilizing glutamic acid, do not appear to be inhibited by the presence of Corexit. Another curious result of one of the experiments (Fig. 45) was that the presence of NH_4^+ enhanced the growth of (glucose utilizing) organisms which were exposed to Corexit. The results of this experiment suggest that the microorganisms were utilizing Corexit for growth and that this process was nitrogen dependent.

During the same experiments, we also measured respiration percentages (Fig. 46 and 47). When the water samples to which only Corexit was added are compared with the controls, the percent respiration was higher. This was not true in the samples treated with glutamic acid where the controls and the Corexit-treated samples produced respiration percentages which were essentially the same throughout the experiment. These data suggest that in natural pelagic microbial populations, Corexit increases the relative amount of labeled substrate that is taken up and respired as CO_2. This metabolic shift may be due to a fixed nitrogen limitation. There are two pieces of evidence that supports this hypothesis; (a) there was very little difference in the respiration percentages observed in the treated and nontreated water samples to which nonlabeled glutamic acid had been added. (b) the water samples that were treated with Corexit and NH_4^+ showed lower respiration percentages than those samples that had not been augmented with NH_4^+.

In summary, the Corexit experiments suggest that the presence of high levels of Corexit alter the composition and/or function of natural pelagic microbial populations; however, it does not appear to reduce the longer term survival and growth of heterotrophic microorganisms capable of utilizing glucose. These data also suggest that the Corexit is actively being degraded by these organisms.

3. Effects of exposing natural microbial populations to both crude oil and Corexit 9527.

We have consistently observed that the addition of Corexit to crude oil in the acute exposure experiments increased the effect to a level greater than would be expected from either one alone. We feel that this indicates an additive stress condition.

4. Short-term effects and management decisions.

Most of the effects of Corexit 9527 that we have observed are probably relatively unimportant in pelagic microorganisms associated with a well-mixed water mass. With the possible exception of the bacteriovorous activity of protozoa, most of the stress effects associated with addition of Corexit to the system would be greatly reduced by the introduction of new species from other waters. In water massess where there is little mixing, a reduction in the diversity of the population could cause long-term changes in microbial function.

Essentially the same could be said for crude oil effects in the water column. If the crude oil becomes incorporated into the sediments, however, we know that many microbial functions will be altered for a significant period of time. For these reasons, efforts should be made to reduce the risk of allowing crude oil to be spilled on water in which there is little net mixing or transport. In the case of an oil spill, the use of the dispersant Corexit 9527 should be restricted in waters with little mixing or net transport.

B. Long-term effects of crude oil on microbial function.

1. Role of bacteria in marine systems - an overview.

Before the effects of crude oil on marine microorganisms can be fully evaluated, one must understand the roles of these organisms in the marine system and the relative importance of these roles in the overall productivity of the system. The main functions or roles of marine bacteria are the following:

a. Bacteria form the major link between primary and secondary production. This works in two ways: bacteria package carbon produced by the primary producers in a form that eventually feeds essentially all of the consumers at all trophic levels. Also, bacteria convert organic nitrogen and phosphate to the inorganic forms required by algae for new plant material.

b. Bacteria control most of the biogeochemical processes which directly affect the productivity of the system. These processes control the concentrations of NH_4^+, NO_3^-, NO_2^-, PO_4^{-3}, H_2S, H_2, O_2, CO_2, methane, and many other important chemical species. The rate at which these compounds are converted from one form to another under a given set of circumstances will often determine the chemical environment at a given point. Because of the reduced diffusion and mixing rates found in marine sediments relative to the water column, the control of the environment by bacterial processes is of particular importance in the sediments.

c. Bacteria can produce ectocrine compounds that may be required for growth of other organisms. These "growth factor" requirements may be particularly important to phytoplankton productivity under certain conditions.

d. Bacteria and blue-green algae (now also classified as bacteria) are the only organisms that are capable of fixing atmospheric nitrogen into a form that is useable by all living organisms.
On a global scale, bacterial transformations are also the most important means of converting fixed nitrogen back into atmospheric nitrogen.

2. The role and relative importance of bacteria in the detrital food chain.

Before proceeding, we would like to define the "detrital food chain" or web. This is the movement of all organic carbon (organic nutrients) from one trophic level to the next which originated from sources other than the direct ingestion and direct utilization of plant material. Thus the nutrients that form the basis for the detrital food chain includes all organics which are not associated with living plant material that is eventually utilized directly by some consumer. Up to a few years ago, the bacterial processes were relegated to a dead-end box at the bottom of flow diagrams which suggested that all of the organic material that could not be used directly by higher trophic levels was mineralized by the bacteria and thus lost from the system. With renewed interest in this problem and the availability of new techniques, much more has been learned about the mechanisms involved in the detrital food chain. As we will show below, the bacteria do indeed mineralize organic compounds; however, they also fill a very important role in the overall productivity of the system that has not become fully appreciated until recently.

In an extensive study of the role of the detrital food chain in juvenile salmon production Naiman and Sibert (1979) stated the following: "The results of our study support the concept of Mann (1972) and Pomery (1974) that the ocean's food web is detrital". Pomery suggested that the main route for the food web in the marine environment was plants - microorganisms (primarily bacteria)-consumers. This is in contrast with the classical view of algae-herbivores-larger consumers, where the bacteria act only as mineralizers.

Mann (1972) concluded that microorganisms provided the main link in the detrital food web while studying the chemical composition of plant material during decomposition. The same conclusion was reached by Thayer et al. (1977) during a similar study of eelgrass decomposition. In a 1975 study, Harrison and Mann showed that the cropping of bacteria by protozoa increased the decomposition rate of eelgrass leaves. Thus, the efficient degradation of this plant material required not only microbial colonization but the cropping of that population by higher trophic level consumers as well.

In the classical scheme, the majority of organic carbon produced by phytoplankton was consumed directly by zooplankton. Recent studies (Smith and Wiebe, 1976; and Larrson and Hagstrom, 1979) have shown that a significant portion of this carbon may be released as soluble carbon and that this in turn, is converted to particulate carbon by bacteria. Paerl (1978) has shown that bacteria colonize the surface of actively growing phytoplankton (over 50% showed colonization). This suggests that the bacteria are utilizing the nutrients being released by actively growing phytoplankton. Using radioactive tracer techniques, Cole and Likens (1979), concluded that bacteria are the major agents of phytoplankton decomposition. In a recent study by Fuhrman and Azam (1980), natural rates of bacterial biomass production were estimated in water samples collected from very different coastal marine locations. They concluded that bacterial biomass production was a quantitatively important component of coastal marine food webs.

As has been shown above, bacteria are important to many aspects of the detrital food chain. They are the most efficient organisms known in taking up and utilizing soluble organic compounds. It has been estimated that if there were no bacteria in the water column, that the level of soluble organic compounds would be at least 100 times greater than that currently found in marine waters. This means that these soluble organics are converted to particulate organic carbon in the form of bacterial biomass which can then be used as a food source for higher organisms. Bacteria also produce enzymes which hydrolyze such recalcitrant compounds as cellulose, chitin and lignin. The resulting soluble compounds are taken up by bacteria and converted to bacterial biomass. The detrital material is colonized by bacteria which is then ingested by higher organisms. The bacteria are digested off the detrital material and detrital particles are then released back into the environment as fecal pellets. These pellets are recolonized by bacteria and reingested. The end result of this process is a food source which has low C:N:P ratios and thus is an excellent balanced organic nutrient. The enrichment of both nitrogen and phosphorous is made possible by the extreme efficiency with which bacteria are able to take up these elements.

In a 1977 review article, Fenchel and Jorgensen presented evidence that quantified the relative importance of the detrital food chain in nutrient transfer through the system. They showed that only 10% of macrophytic and seagrass biomass was grazed on directly by herbivores. Considering the composition of most terrestrial carbon, we assume this would hold true for this potential nutrient source as well. If the estimates that over 50% of the phytoplankton produced carbon is also routed through the detrital food chain are correct, the total nutrient flow via this route is substantial. Fenchel and Jorgensen (1977) estimated that on the average 50% of all primary productivity is utilized via this route in the worlds oceans. The figure for nearshore environments runs near 80%.

3. The role and importance of bacteria in the mineralization process.

During the mineralization process, the carbon, nitrogen, phosphorous, sulfur and trace elements that are incorporated into organic molecules are converted into inorganic forms that are then reincorporated into organic molecules by biological activity (primarily through the actions of plants). The high metabolic activity found in bacteria, make them the most important factor in this process. In systems in which fixed nitrogen is limiting for phytoplankton growth, the rates at which bacteria are capable of mineralizing organic material may directly affect the rate of primary productivity by phytoplankton. During a seasonal study of relative microbial activity in waters near Vancouver Island, B.C., a pulse of high microbial activity was observed both before and after the major spring phytoplankton bloom (Albright, personal communication). It was felt that the first pulse in microbial activity might have released the inorganic nutrients required for the spring bloom. Schell (1974) reported that there was strong evidence that microbial mineralization processes were the most important factor in the accumulation of inorganic nutrients during the winter months in the Beaufort Sea. It was suggested that this was the source of inorganic nitrogen required for the spring phytoplankton bloom.

4. The role and importance of microbial nitrogen fixation in the marine environment.

In recent years, a new, sensitive, fast and relatively inexpensive assay for measuring _in situ_ rates of nitrogen fixation has been developed (Stewart, 1967). This has greatly facilitated the study of this process in both terrestrial and marine systems. At this point there have been a number of studies which have reported rates of nitrogen fixation in marine sediments. Rates that have been reported in colder marine environments compare well with those that we have observed in our studies. There is, however, a great deal of difficulty in determining what significance this has to the total nitrogen budget of a given body of water. In the last three years, there have been reports of studies where the relative importance of nitrogen fixation to the nitrogen requirements of specific marine plant species have been estimated (Capone et al., 1979; Teal et al., 1979; and Zuberer and Silver, 1978). In all three cases, nitrogen fixation was thought to account for about 30% of the nitrogen requirements for the plants studied.

Although we do not have the information required to report a complete nitrogen budget for the areas we have studied, we do have indirect evidence that nitrogen fixation may be important to the function of the detrital food chain in marine sediments. It is generally accepted that nitrogen is limiting for carbon utilization in many marine sediments. This would be particularly true in sediments where the primary organic carbon input was low in organic nitrogen; i.e., macrophytic and terrestrial carbon. During cruises in Cook Inlet, Shelikof Strait, and Norton Sound, we measured nitrogen fixation rates while Dr. Atlas and his associates measured natural rates of denitrification. Thus, the net fixed nitrogen generated in the sediments was compared to the removal of fixed nitrogen to atmospheric nitrogen by denitrification. In Kamishak Bay, the mean rate of denitrification was much greater than the mean rate of nitrogen fixation. This indicated to us that detrital particles generated in the Lower Cook Inlet during a period of phytoplankton blooms were probably settling out of the water column and into the sediments. Nitrogen rich detrital particles from Upper Cook Inlet may also contribute to the organic content of Kamishak Bay sediments. This transport would probably bring in exogenous organic nitrogen which could be mineralized (ammonification) and oxidized (nitrification). The end result of these processes would make more nitrate available for denitrification. In addition, the presence of organic nitrogen would inhibit nitrogen fixation.

We know that currents on the west side of Cook Inlet travel south into the Shelikof Strait. As detritus that is generated in Cook Inlet is carried south by this current, we would anticipate that the nitrogen content would be decreased as the available organic nitrogen is used up. The bacteria associated with the detritus should preferentially utilize organic N-containing compounds (mostly protein). The bacteria would then be cropped, thus removing the nitrogen from the particles. The end result of these processes would be the deposition of detritus material with high carbon to nitrogen ratios, i.e. low concentrations of organic nitrogen. If this were the case; one would predict that the rates of nitrogen fixation would be relatively high and should approximately equal the rate of denitrification. This is exactly what was observed in Shelikof Strait. The mean value for nitrogen fixation was higher in the Shelikof Strait than in any other region that we have studied (see Section I for details). These data suggest to us that nitrogen fixation is an important and constant feature of the detrital food chain in the Shelikof Strait.

A similar pattern was observed on a seasonal basis in our 1979 Kasitsna Bay studies. In the sediments near Kasitsna Bay, there was a major input of carbon into the sediments during the summer which was reflected in much higher benthic relative microbial activity than at any other time of the year. It was during this same season that we observed the lowest mean value for nitrogen fixation. The highest mean value was observed in November. We feel that the detritus that settles out of the water column during the late spring and summer contains a relatively low carbon:nitrogen ratio; thus the low nitrogen fixation rates. As the bacteria preferentially utilize the compounds containing relatively high concentrations of nitrogen and this nitrogen is removed as the bacteria are cropped, the carbon:nitrogen ratio in the detritus increases; i.e., there is a net loss of fixed nitrogen. This results in high rates of nitrogen fixation in the late fall when there are still relatively large amounts of easily utilizable carbon available but very little fixed nitrogen. As the season progresses, the nitrogen fixation rates decrease because carbon in a readily available form becomes depleted.

One other observation leads us to conclude that nitrogen fixation might be important to the detrital food chain. Using the mean yearly rate of nitrogen fixation in the sediments near Kasitsna Bay, we calculated that the concentration of all forms of fixed nitrogen in the interstial waters of the same sediments could be replaced by nitrogen fixation every 24 hours.

5. Long-term crude oil effects on microbial function in marine sediments

a. Overview

   1. Changes related to the detrital food chain perturbation.
      a. Decreased bacterial cell biomass.
      b. Decreased microbial activity, increased respiration percentages and therefore reduced microbial biomass production.
      c. Decreased total adenylates (decreased total biomass).
      d. Decreased nitrogen fixation rates.
      e. Increase in detritus accumulation on sediment surface.
      f. Decreased infaunal activity such as burrowing.
      g. Decrease in structural polysaccharide degradation (enzyme activity)
   2. Changes related to mineralization.
      a. Decreased alkaline phosphatase activity.
      b. Decreased microbial activity.
      c. Decreased denitrification and nitrogen fixation.
      d. Decreased oxygenated zone and extension of this zone via infaunal burrowing.
   3. Changes related to altered chemical environment.
      a. Increase in hydrogen ion concentrations in surface sediments.
      b. Decreased redox potential in surface sediments (anoxic conditions prevail).

c. Presumed increase in hydrogen sulfide concentrations.

d. Increase in methane and CO_2 concentrations.

b. Crude oil effects on the detrital food chain.

There now exists in the literature, extensive documentation of the effects of crude oil on benthic organisms (Boucher, 1980; Carr and Reish, 1977; Elmgreen et al., 1980; Giere, 1979; Swenmark, 1973; and Taylor and Karinen, 1977). If these organisms are killed or driven out by the presence of crude oil, several important changes will take place in the system. The burrowing activity, which is now known to have an important function in the overall metabolism of the sediment by increasing the total oxidized surface area, is reduced. Burrowing activity is also responsible for the turning over of the sediments (more will be said about this in the next sub-section). Of greater importance to the detrital food chain is the interruption in the flow of bacterial biomass which is utilized by higher trophic levels. If there are no organisms present that are capable of cropping the bacteria, then the sediments become in effect, carbon sinks. The organic carbon that would normally be used as the basis for most secondary productivity will remain in the sediments and will not be used as a food source for the rest of the food chain.

Our study of oiled sediments indicates that this is what is taking place. The non-oiled sediments showed very little accumulation of detrital material even at the height of the spring phytoplankton bloom. They also contained the normal 2-3 cm of oxidized sediment on the surface and extensive evidence of infaunal borrowing. In contrast to this, the oiled sediments had extensive detrital deposits on the surface and there was essentially no oxidized layer on the surface of the sediments. In addition, there was no evidence of burrowing activity.

Our measurements of total adenylates indicate that there was reduced total biomass in the oiled sediments. This reduction was almost twice that which would be expected from the known reduction in bacterial biomass as determined by direct counts (epiflucrescent microscopy). We therefore conclude that the balance of the reduction was due to reduced infauna which are potentially bacteriovorus.

There have been a number of studies in which it has been shown that microbial activity is actually stimulated when bacteria are being actively cropped (Harrison and Mann, 1975 and Fenchel and Jorgensen, 1977). We have observed both a reduction in microbial activity and a reduction in bacterial biomass in oiled sediments. This could, in part, be due to the absence of bacteriovores in the oiled sediments. It is more likely; however, that this reduction is in response to the toxic effects of the crude oil itself. We have shown that microbial activity is reduced in the presence of crude oil under short-term (8 hour) exposures. In addition, we have seen that the respiration percentages are increased in presence of crude oil. This means that proportionately more of the nutrients that are utilized by the bacteria are being respired as CO_2 and less is being incorporated into bacterial biomass. Even if the bacteria were being cropped, this would mean a greatly reduced effeciency in converting detrital carbon into usable carbon for secondary productivity.

Another indication of stress induced by the toxic properties of crude oil is the decreased energy charge ratios observed in oiled sediments. These ratios, which are generated by comparing relative levels of ATP, ADP, and AMP, are an indication of the metabolic activity of microorganisms (Wiebe and Bancroft, 1975). The higher the ratio, the more active the population; conversely, the lower the ratio, the less metabolically active the population.

An additional major function of microorganisms in the detrital food chain is to produce enzymes which breakdown organic compounds which would otherwise accumulate in the sediments and thus would not be utilized by higher trophic levels. Our studies have shown that crude oil reduces the activity of some of these enzymes. More specifically, we have observed that the enzymes that breakdown cellulose and chitin (two structural polysaccharides) are inhibited (Table 45 and 46). Actually, our data suggests that the enzyme itself may not be inhibited, but that the formation of the enzyme and/or the growth of the organisms that produce these enzymes are inhibited. Regardless of the mechanism involved, the results of our effects studies suggest that the actual degradation of chitin (the structural material in invertebrate shells), cellulose (the structural material in plants) and dried macrophytes is reduced in the presence of crude oil. Our data further suggests that the change in degradation rates of these compounds may account for changes that we have observed in many of the other variables that we have been monitoring.

Under normal conditions, these enzymes would solubilize these complex compounds into simple moelcules which are, in turn, taken up by the bacteria to form new biomass. This biomass would then be utilized as a food source by higher trophic levels. It appears from our data that crude oil interferes with this process at all of these levels; from the enzymes that breakdown the complex organic molecules, to the formation of bacterial biomass, to the utilization of bacterial biomass as a food source.

It should be pointed out that during the course of the same studies, we observed that crude oil apparently stimulated the enzyme activities of those enzymes that breakdown storage polysaccharides such as starch and algin. These are storage products that are found in phytoplankton and macrophytes, respectively. It is thus very possible, that the effect of crude oil on nutrient cycling may depend on the types of organic compounds found in the impacted area. Although we have no direct information about what the impact would be in sediments where the primary nutrient source is from phytoplankton, our studies with a dried macrophyte suggest that the breakdown of this material would be adversely affected by the presence of crude oil.

c. Crude oil effects on mineralization and nitrogen fixation.

During our studies, we have observed changes due to the presence of crude oil that strongly suggest that the rate of mineralization is reduced. These effects could have a direct bearing on primary productivity rates of marine algae. We have direct evidence that mineralization rates for simple soluble organics can be reduced by over 50% (the glucose, glutamic acid and acetate uptake rates shown in Tables 27-31). These reduced rates may, in part, be related to the reduced oxygenated zone observed in the oiled sediments. Most normal marine sediments contain two zones; a top oxidized zone in which most of the mineralization of organic material by microorganisms takes place and an anaerobic zone beneath it (Vanderborght et al., 1977). In a recent review of mineral cycling by Fenchel and Blackburn (1979), it was shown that the presence of the top oxygenated zone is essential for normal mineralization of organics. Without this zone, the products of anoxic metabolism can escape from the sediments. Many of these products are toxic to most higher (eucaryotic) organisms. Under most conditions, more than 90% of all mineralization takes place in the oxygenated zone near the sediment surface. Our studies of redox potential changes suggest that this zone is essentially eliminated in oiled sediments.

Classical studies of marine sediments often treat sediments as a two dimensional layered system. Recent studies have shown that this model is a gross oversimplification of the actual condition in marine sediments (Rhodes and Young, 1970 and Gerlach, 1978). For example, the burrowing activity of infauna can greatly increase the extent of the oxidized layer mentioned above, it can act to physically turn over the sediments bringing to the surface nutrients that would be left at depth and the pumping action by some organisms can bring oxygen and oxygenated compounds into the deeper sediments. Dennis Lees (personal communication) has calculated that the burrowing activities of one species of clam would increase the surface area in Western Cook Inlet sediments by roughly 2.5 times. This is only one of many species present which could provide this function. If crude oil eliminates key infaunal species, the rates of mineralization would certainly be reduced even if there were no other detrimental effects.

In addition to the general reduction in mineralization rates, we also have conducted observations that suggest that specific reactions which are involved in mineralization are affected by the presence of crude oil. These effects would be additive to those described above. We have observed that the natural rates of

denitrification were reduced below detectable levels in sediments exposed to crude oil for periods of up to one year. This represents at least a 97% reduction in activity. When the same experiments were conducted in sediments to which nitrate was added, the crude oil treated sediments showed a 57% reduction in denitrification rates. The denitrification reaction converts nitrite to nitrous oxide and then to atmospheric nitrogen. If an excess of nitrate is added to the system, this is converted to nitrite which is then utilized in the above reaction. In the non-augmented samples, the activity in oiled sediments was reduced to less than 3% of the control. This is in contrast to a rate that was 33% of the control when nitrate was added. It thus appears that the reaction that converts ammonium ion to nitrite may be impaired in the oiled sediments. Recent measurements of denitrification in Elson Lagoon oiled sediments by Dr. Atlas have shown similar results. In his study, there was no difference between oiled sediments and the controls in experiments where nitrate was added but there was a significant reduction in natural rates (see his final report for RU #29).
We have not been able to document by direct observation the effects of crude oil on ammonia oxidation because we have not been able to detect this process in Kasitsna Bay sediments.

Another function of microorganisms is the conversion of organic phosphate to inorganic phosphate. The enzyme that is responsible for this reaction is phosphatase. In both the oiled sediments from Elson Lagoon and Kasitsna Bay, we have consistently observed lower phosphatase activity in these sediments relative to the controls. We have also observed that crude oil reduces the activity of the enzyme arylsulfatase. The exact function of this enzyme in marine sediments has not been documented but it has been suggested by Oshrain and Wiebe (1979), that this enzyme may initiate arylsulfate ester metabolism. Thus, this enzyme may be important in mineralizing components of marine algae.

Another microbial process that adds nitrogen to the system is nitrogen fixation. In the Kasitsna Bay, we have consistently observed that the rates of nitrogen fixation in oiled sediments are greatly reduced relative to the controls. This effect takes place fairly rapidly (within a few days) and it occurs at relatively low crude oil concentrations.

d. Crude oil effects on sediment chemistry.

We have observed several phenomena which suggest that the chemical properties of sediments are altered by the presence of crude oil. These changes undoubtedly reflect changes in microbial activities caused by the crude oil. There is a marked reduction in the redox potential in the surface of the sediments. This is probably due to the reduction in the available O_2 present. These anaerobic conditions are also reflected in the increased rates of methane production that we have observed in oiled sediments. Although we have not measured it directly, there is strong presumptive evidence that another product of anaerobic metabolism, hydrogen sulfide (H_2S) is also being produced. The human nose is very sensitive to hydrogen sulfide (rotten egg smell). Our noses have consistently detected H_2S in oiled sediments. In addition, we have observed the growth of bacteria known to utilize H_2S on the surface of oiled sediments but not on the surface of the controls.

We have also observed changes in the sediment surface hydrogen ion concentration (pH) during the first 5 months of exposure. The hydrogen ion concentration increases (pH decreases) in the oiled sediments. With exposure periods beyond 5 mo, these differences were no longer statistically significant. Under these acidic conditions, there should be a significant shift in the chemical balance of a number of potentially important chemical species (Fenchel and Blackburn, 1979)

These data suggest that crude oil causes significant changes in the sediment surface which will undoubtedly effect recruitment of benthic organisms into the impacted area. These effects may be present long after the initial toxicity of the crude oil has dissipated.

e. The effects of "weathered" crude oil and crude oil overlays

All of the effects that have been described above have been observed in sediments that had been thoroughly mixed with fresh crude oil. We conducted one experiment in which sediments were exposed to "weathered" crude oil at two concentrations for a period of 1 year. The variables that were measured were glucose uptake and respiration, phosphatase activity, redox potential, and nitrogen fixation showed the same changes that we observed in fresh crude oil exposures except in some instances the effect was somewhat less. The nitrogen fixation rates, on the other hand, did not show a significant change from the controls when exposed to "weathered" crude oil. This suggests that nitrogen fixation may be inhibited by some volatile fraction of crude oil. Knowles and Wishart (1977) reported observations that seem to substantiate our hypothesis. They found that Beaufort Sea sediments that had been exposed to "weathered" crude oil, hexane, decane, dodecane, and hexadecane had no effect on nitrogen fixation; however, 1, 2, 4 trimethylbenzene completely inhibited nitrogen fixation in the same sediments.

During a study of crude oil impact on soft marine sediments following the Tsesis oil spill, Elmgren et al. (1980), conducted hydrocarbon analyses on the sediments in the impacted area. They concluded that most of the crude oil that settled out of the water column became associated with the flocculant layer at the sediment-water interface. Although we were not able to duplicate these conditions, we did attempt to approximate this situation by placing a layer of crude oil treated sediment approximately 1 cm thick over a nontreated sediment. This experiment was conducted using both fresh and "weathered" crude oil. Under these conditions, the fresh crude oil produced statistically significant changes which were essentially the same as those observed in the experiments in which the entire sediment was exposed to crude oil. The only change that was statistically significant in the "weathered" crude oil overlays was a reduction in denitrification rates of 72%. These observations suggest that the actual effects of crude oil on microbial function under spill conditions may depend on the degree of weathering and the way in which the crude oil is mixed with the sediments.

f. The effects of fresh crude oil on microbial function in sediments augmented with organic nutrients.

A series of experiments were conducted on Kasitsna Bay sediments in which a number of different organic substrates were added. These experiments were designed to provide information about how crude oil might affect microbial function when various types of organic nutrients were present and to determine how the presence of two of these substrates (starch and chitin) would effect crude oil degradation. The results of the degradation data are presented in Section V of this report.

The interpretation of these data is complicated by the fact that there are so many variables that must be considered. For example, if crude oil inhibits the breakdown of a given substrate, then we would expect to find a lower O_2 demand and less energy to drive such energy requiring reactions as nitrogen fixation. If nitrogen containing substances are broken down, then we would expect to see an increase in ammonia which may be toxic at higher concentrations to both nitrogen fixation and glucose uptake. In summary, while interpreting these results, changes in ammonia, O_2, and available carbon must all be considered. It also appears that exposure time might also change the observed effects.

1. Effects on denitrification potentials.

Regardless of the substrate added, there was always a depression in potential denitrification rates when the nontreated and substrate augmented sediments were compared. The same can also be said of redox potentials indicating that the substrate is being oxidized with a resulting reduction in available O_2 and other potential electron acceptors. In most cases, when fresh crude oil is added to the system, the denitrification rates are further reduced. There were three cases in which this was not true. In two studies, there were significant increases in denitrification potential in oiled sediments supplemented with cellulose. The other exception was under the same conditions when the sediment was amended with chitin. During the amended tray study, we also observed that the enzymes that breakdown chitin and cellulose were inhibited by the presence of crude oil. Thus in the presence of crude oil, we would expect that fewer of the breakdown products from these two compounds would be available to the microbial populations in oiled sediments amended with these substrates. If fewer breakdown products are available for oxidation by the microorganisms, then we would expect that the redox potentials would not be as low under these conditions. This is exactly what was observed. From these data, it would appear that the reductions that we observed in denitrification rates in the presence of crude oil was due to the secondary effect associated with reduced redox potential rather than a direct toxic effect.

2. Effects of crude oil on nitrogen fixation rates.

There were only two cases where nitrogen fixation rates were not reduced in the presence of added substrates. One where cellulose was added and one where starch was added. It is not known why a reduction did not occur in this one instance with cellulose. In the other instance, we strongly suspect that the observed stimulation was caused by a contamination of the starch with a simple sugar.

There were four cases where nitrogen fixation was stimulated in the presence of crude oil in augmented samples; twice when starch was added and twice when chitin was added. Since we have shown that amylase is stimulated in the presence of crude oil, the increase in nitrogen fixation in the starch ammended sediments could be due to the increase in glucose production which provides the energy source for nitrogen fixation. Under these conditions, there would be a shortage of fixed nitrogen which would optimize the conditions for nitrogen fixation. These ideal conditions would offset the toxic effects of the crude oil. The resultant nitrogen fixation were as high or higher than the nonamended controls.

In the sediments amended with chitin, the slight increase in nitrogen fixation rates in those samples exposed to crude oil could be due to the inhibition of chitobiase. The breakdown of chitin would release fixed nitrogen, probably in the form of ammonia, which would tend to inhibit nitrogen fixation; a phenomenon that is well documented in the literature.

In summary, these studies suggest that crude oil can cause both direct toxic effects and indirect effects that depress nitrogen fixation. The studies with "weathered" crude oil suggest that the toxic effects are caused by components in the volatile fraction of fresh crude oil. These studies also suggest that the reduction in redox potential is not the secondary or indirect effect that is causing the reductions in nitrogen fixation observed when organic substrates are added to the sediment.

3. Effects on glucose uptake and respiration.

During the course of our Kasitsna Bay studies, we have measured the effect of reducing available O_2 in glucose uptake experiments. We have found that there is little effect on glucose uptake but there is always an increase in the respiration percentage. In every case where the sediments were supplemented with organic substrates, the respiration percentage increased. Under more anaerobic conditions, the efficiency of energy production per mole of substrate decreases, therefore one would expect to find proportionately more substrate being mineralized to provide a given amount of energy. A careful analysis of the redox data shows; however, that the changes observed in oiled sediment percent respiration is not entirely caused by a change in available electron acceptors. For example, in the cellulose and chitin amended trays, the redox potentials changed very little or actually increased in the presence of crude oil. All of these sediments showed increases in respiration percentages when compared to the amended controls (three out of four showed significant changes).

When we look at the glucose uptake data, it is apparent that redox potential has very little effect on this function. In most cases, when the sediment was amended with an organic substrate, the glucose uptake rates increased. When crude oil was added to these sediments, the uptake rates decreased in most cases. Where there was little or no change, secondary effects which countered the toxic effects of crude oil were the apparent causes; i.e., in the case with starch where crude oil stimulated the production of glucose. These observations along with the results of the short-term crude oil effects studies suggest that the primary crude oil effect on glucose uptake is direct.

4. Effects on CO_2 evolution and methane concentrations.

The results of these measurements are very difficult to interpret for a number of reasons. First, the degradation of crude oil most likely produces the increase in CO_2 production. But as the redox potential is reduced, one would expect to find a higher proportion of carbon released as methane as conditions become more anaerobic. Our time-course studies have shown that upon initial exposure, crude oil depresses the formation of both of these compounds, but as the exposure time is increased, the production of these gases increases as the crude oil is degraded and the sediments become more anaerobic.

5. Summary of amended sediment observations.

These studies have shown that the primary effect of fresh crude oil on nitrogen fixation and glucose uptake is a direct-toxic effect. On the other hand, the primary effect on denitrification and CO_2 production and methane concentrations is probably indirect. The effect on denitrification is closely linked to the resulting redox potential. The effect on respiration percentages is both primary and secondary.

g. Effects of crude oil on microbial function in sediments collected from various locations.

We conducted observations on the effects of crude oil on microbial function in both sediments collected from the Beaufort Sea (Elson Lagoon) and the Cook Inlet (Kasitsna and Coal Bays). The variables that were affected by crude oil in one location were usually affected in the other as well. The exception to this was nitrogen fixation. Even after 2 years exposure, we did not observe a significant reduction in Elson Lagoon sediment nitrogen fixation rates in crude oil treated sediments. This suggests that although most of the changes in response to crude oil perturbation are essentially the same in Arctic and sub-Arctic marine sediments, some basic differences do exist.

When we compared the measurements made in Kasitsna Bay and Coal Bay sediments, we also noted differences; most of these were quantitative in nature. The only major difference observed was in CO_2 production rates which were depressed instead of being increased.

6. Long-term effects of the dispersant Corexit 9527 on microbial activities.

During our Kasitsna Bay studies we have observed the long-term effects of Corexit on a number of microbial functions. At a concentration of 500 ppm, the only effect observed was that expected from the addition of an organic nutrient to the sediment. These observations did not; however, include measurements of bacterial species diversity. We did, however, observe increased effects when Corexit was added to crude oil. The Corexit appeared to enhance the toxic effect of the crude oil in the initial months of exposure. This same phenomenon has also been reported by others who have studied other marine organisms (Swedmark et al., 1973).

C. Seasonal observations of microbial function in the waters and sediments near Kasitsna Bay.

One of the objectives of our Kasitsna Bay work was to determine what seasonal changes might occur in certain key microbial functions. These observations were designed to accomplish several objectives; (1) to determine natural seasonal variation, (2) to determine the major sources and sinks of carbon and nitrogen in the system, (3) to determine if there were seasonal differences in the actual or potential impact of crude oil on key microbial processes, and (4) to determine the relative importance of various microbial processes which might be impacted by crude oil perturbation.

1. Observations on the carbon cycle.

By analyzing the relative microbial activity in waters and sediments, we have an indirect measurement of the organic carbon that comes into the system. These measurements along with primary productivity determinations show that there is a major input of organic carbon from the water column in either late spring or early to mid summer. In 1979, there was a major phytoplankton bloom in April with a secondary bloom in August. In 1980, some phytoplankton growth was observed in April but most of it occurred in June with a secondary peak in August. There was apparently a lag between the input of this carbon and changes in the microbial activity in the sediments which would reflect the presence of this carbon. The peak in relative microbial activity that we observed in the sediments in August, 1979 might not, however, have been due to the input of carbon from phytoplankton but from the summer input of terrestrial carbon and carbon from macrophyte biomass. Although there is no quantitative data available on the magnitude of the terrestrial carbon input into this region, it is known that there is considerable macrophytic production at this time (Lees, personal communication). If the major input of organic carbon into the sediments is from these sources, the presence of crude oil could have a very severe impact on the detrital food chain in this and similar regions since it is known that most of this material must be converted to bacterial biomass before it can be utilized by the rest of the food chain. In addition, we have shown that crude oil adversely affects the enzymes that would be primarily responsible for the breakdown of their structural components.

Our studies of relative microbial activity in the sediments located near Kasitsna Bay indicate that terrestrial input represents a major portion of the carbon coming into the sediments at the heads of the two major bays in the region. During all field studies, the relative microbial activity found in the sediments near the head of these bays was significantly higher than the locations toward the mouth.

The study of relative microbial activity and phosphatase and arylsulfatase activities in the sediments of Kachemak Bay indicate that most of the carbon present in this bay may come from marine sources. We do not know at this time if this material is primarily from phytoplankton or from macrophytes.

2. Observations on the nitrogen cycle

Although rates of denitrification were generally below the detection limits of the techniques used, we were able to detect relatively high rates of nitrogen fixation in the sediments near Kasitsna Bay. On a seasonal basis, we observed significant changes which appear to be linked to the input of organic substrates. The patterns observed in this study suggest that nitrogen fixation is linked to the demand for fixed nitrogen, when the availability of fixed nitrogen in the remaining detritus is reduced. Thus, the initial effect of crude oil on nitrogen fixation would probably be of lesser importance in the late summer than at any other time of the year. Although, as we have seen in the long-term effects studies, the duration of the adverse effect of crude oil on nitrogen fixation is long enough that the season at which the initial impact takes place is probably relatively unimportant.

D. The importance of long-term crude oil effects relative to overall productivity.

With the information that is available to us at this time, it appears that in areas where levels of fresh crude oil reach 1 ppt, there would be a measurable reduction in the numbers of all organisms present that are dependent on food sources in the impacted area. On Kachemak Bay and similar bays in the sub-Arctic marine environment, the total amount of available food cycled through the detrital food chain should range from 50 to 90%. Our studies on crude oil effects indicate that a concentration of 1 ppt, the production of bacterial biomass (the basis for this food chain) is reduced by a least 50% in sediments exposed for as long as one year. Even if this was the only adverse effect caused by crude oil, we would expect to find a reduction of 25 to 45% in the amount of food available to the higher trophic levels in the impacted. area. Since it is known that many commercially valuable species in Kachemak Bay are directly dependent on food from the detrital food chain (Feder, final report for RU #5), it is evident that an extensive spill could have a serious impact on the commercial fishing industry in this region.

At this time, we have no good estimate on how much of the inorganic nutrients present are generated locally by microbial mineralization. If this is an important source of inorganic nitrogen and phosphorous, for primary production, we would anticipate a further reduction in overall productivity. Another unknown at this time, is the extent to which crude oil affects the organisms that are the primary detrital feeders in the system. Any adverse effect on them would also reduce the overall productivity of the system. Thus, it can be seen, that if crude oil does become incorporated into these marine sediments, our estimation on the effects of crude oil on this system is probably low.

The actual impact of a major spill to the overall productivity of the impacted area would depend on a great number of variables. Some of these include the type, degree of weathering and concentration of oil incorporated into the sediments. It would also depend on how thoroughly the oil was mixed with the sediments and the extent of the impacted area. In addition, it would also depend on the relative importance of the area to the juvenile stages of key species. It is obvious that much more work remains to be done before a realistic estimate of potential environmental impact can be made for a given set of circumstances. The duration of the impact is another factor that must be kept in mind. In the introductory statement made by Vandermeulen (1978) during a symposium on "Recovery Potential of Oiled Marine Northern Environments", he suggested that it might take 5-15 years before the oil impacted region would return to normal. In a long-term study of the effects of #2 fuel oil on a section of a Massachusetts coasline impacted following a spill, Sanders et at. (1980) reported that there were effects on the biota that were still measurable after five year.

In an attempt to make some projection as to the duration of the crude oil effects in the Kasitsna Bay tray study, we have plotted percent reductions in glucose and glutamate uptake rates and nitrogen fixation for the five sampling periods (data taken from Tables 27-31). Omitting the one low observation for glutamate uptake, the points were fitted alternately to a linear ($Y = a + bX$) and a power curve $Y = aX^b$) reduction, we can estimate possible maximum and minimum times for the specific crude oil effects to disappear. The extrapolated values were: glucose uptake - 2.6 to 6.8 years, glutamate uptake - 1.6 to 3.3 years, and nitrogen fixation - 1.9 to 8 years. However, it must be noted that this current study was not continued long enough to accurately determine the actual rate at which the crude oil effects will subside.

In the Elson Lagoon study, measurable changes in many variables did not occur until the sediments had been exposed for 9 months. Once these changes were initiated, they persisted for at least 2 years (the length of the study). The results of our studies suggest that the effects of crude oil in the sediments of the Beaufort Sea would persist for a much greater period of time than that observed in the more temperate climate of Cook Inlet. Judging by the delayed onset of measurable change, the impact could last up to 10 times longer in the Beaufort Sea. This is probably due to the length and degree of metabolic depression during the winter months in the Beaufort Sea inshore sediments.

E. The relevance of our observations to actual spill conditions.

We designed our experimental conditions so that the changes that we observed would be similar to those observed under conditions of maximum impact; e.g., we thoroughly mixed fresh crude oil directly into the sediments at relatively high concentrations. We did this in order to make sure that we would be able to measure changes that might take place. As it turned out, the observed changes were so dramatic that this measure may not have been needed. Although the concentration used in these experiments seemed high at the time we initiated this experiment, a recent report by Vandermeulen et al. (1979) showed crude oil concentrations twice this magnitude in sediments along the coast of France following the Amoco Cadiz spill (110 ppt). It would thus appear that the concentration that we used in the initial experiment was high but yet possible under actual spill conditions.

More realistic levels of petroleum hydrocarbons in marine sediments would be in the range of about 0.1 to 1.0 ppt in heavily contaminated sediments following an oil spill (Boucher, 1980). Although high levels of petroleum hydrocarbons were not found in subtidal sediments following the Tsesis spill, concentrations of up to 7 ppt were observed in the particulate matter that was settling out of the water column (Boehm et al., 1980). In the same report, it was suggested that current sampling methods do not allow the accurate sampling of the flocculant layer at the water-sediment interface which probably contains most of the hydrocarbons.

We conducted a series of experiments that were designed to determine what effects crude oil might have on microbial function at concentrations ranging from 0.1 to 50 ppt. With the exception of denitrification potentials and phosphatase activity, all of the variables that we studied showed the greatest change in function in sediments that had been exposed to crude oil concentrations at or below 1 ppt. Higher concentrations generally had little additional effect. Significant reductions in microbial biomass production, redox potentials and nitrogen fixation rates were all observed at 0.5 ppt. Measurable but not statistically significant changes were observed in the same variables in sediments exposed to 0.1 ppt fresh crude oil. It would thus appear very probably that the types of observations that we have documented would be found under actual spill conditions.

The only data that we have had an opportunity to gather under spill conditions were those obtained from Glacier Bay in the spring of 1979. A small amount of fuel oil was spilled by a cruise ship in Glacier Bay, AK, located north of Juneau. Our analysis of intertidal sediments collected from the spill site indicated that changes did occur in glucose uptake rates and respiration percentages.

The only other observation that has come to our attention that substantiates our findings under spill conditions was reported after the Tsesis oil spill (see final report). They observed that the oxygen concentrations in the bottom waters were depressed in the area of the spill. This is an observation that we would have predicted from our results.

F. Recommendations regarding the types of measurements that should be made during a major spill to assess environmental impact.

During past environmental impact studies of major oil spills, the primary emphasis has been in assessing the direct impact of crude oil on higher organisms and the type and concentration of petroleum hydrocarbons associated with the impacted area. More recently, assessments of crude oil degradation rates and direct effects on marine algae have also been made. The results of our studies strongly suggest that an additional set of variables should also be measured. These would assess the impact on primary and secondary productivity over an extended period of time. The functions that appear to be the most susceptible to crude oil perturbation are microbial biomass production, nitrogen fixation rates and redox potentials. We would therefore recommend monitoring these variables to assist in assessing environmental impacts. These are very sensitive variables that could act as indicators of general shifts in microbial processes. Since crude oil tends to reduct O_2 levels, the concentrations of gases and ions that are associated with anaerobic processes should also be monitored. This would pinpoint areas where the sediment surface chemistry is altered by the presence of crude oil.

Our pilot study of oiled sediments from Glacier Bay suggests that even without baseline data available for comparison, one can show the impact of petroleum hydrocarbons by measuring microbial function. This study did show us however, that "control" (non-oiled) sediments should be collected and analyzed along with the oiled sediments. Care should be taken in selecting "control" sediments with physical characteristics similar to the oiled sediments and the number of non-oiled sediments should approximately equal the number of oiled sediments analyzed.

We would like to emphasize the necessity of planning the sampling procedures, the types of studies to be conducted and the logistics required to conduct an environmental impact study prior to a major oil spill. This was one of the major conclusions of the personnel who were involved in managing the Tsesis oil spill study. In order to study a spill in Alaskan waters, it is imperative that a comprehensive plan be established, if for no other reason than to provide logistics for such a study. Advanced planning would be particularly critical if chemical and microbiological variables were to be monitored; since unlike many other types of observations, such measurements require field analyses of samples that cannot be preserved for future measurements.

VI. Needs for further study

A. The recent Bering Sea synthesis meetings conducted by NOAA have shown that very little information is available concerning the microbiology of the Bering Sea (Eastern Bering Sea Shelf: Oceanography and Resources - in press). During the same series of meetings, it was shown that the area of the St. Georges Basin is one of the most productive and important fisheries in Alaska. In a recent article by Iverson et al. (1979), it was concluded that in the very productive middle shelf region of the ST. Georges Basin, most of the food available to higher trophic levels was routed through the detrital food chain. If significant quantities of crude oil become incorporated into the sediments of this region as a result of crude oil production and transport, it is quite likely that this fishery will be impacted.

During the Bering Sea Ecological Processes Workshop held in July 1980, it was recommended that one of the objectives of future studies in the southern Bering Sea should be to determine the answer to the question "is the growth of benthic crab food a direct function of detrital flux, and if so, what is the rate of conversion of detrital material to benthic food?" This is an objective that cannot be fulfilled without knowledge of the microbial component of that food chain since it forms the basis for it.

We strongly recommend that a study of microbial function, as it relates to nutrient recycling, be conducted in the St. Georges Basin. At this time, the importance of the detrital food chain in this region can only be guessed at by indirect means. A study of this nature would provide more direct evidence for this relationship, if it exists. It is also quite likely that microbial processes provide much of the inorganic nutrients required for the high rates of primary productivity in the region. The presence of high concentrations of inorganic nutrients in the photic zone is caused by wind driven currents that mix the upper portion of the water column with the nutrient rich bottom layers. Since there is very low advective flow in this region, it is quite likely that much of the nutrients found in these deeper waters have been generated locally by microbial mineralization in the sediments. This is also something that needs to be documented.

Our effects studies have shown that crude oil interferes with both the detrital food chain and mineralization processes. Although these effects have been documented elsewhere (Beaufort Sea and Cook Inlet), these effects need to be documented for this region as well. This study could be conducted by collecting sediments from the St. Georges Basin near the end of a cruise and transporting them to the field laboratory at Kasitsna Bay. Once the sediments were taken to Kasitsna Bay, a time series experiment could be conducted using various concentrations of crude oil. By measuring key variables for periods up to one month, we would be able to document long-term effects in these sediments.

B. Although our Kasitsna Bay effects studies were comprehensive in scope, there are additional studies that should be conducted.

These studies would enable us to provide predictive capabilities that do not currently exist. The studies in which we compared "weathered" and fresh crude oil effects showed that there were differences in the observed effect; this was especially true for nitrogen fixation. A series of long-term experiments should be conducted in which the effects of the three major fractions of crude oil (polar, aromatic, and aliphatic) on microbial function at various concentrations are followed. This would enable us to predict qualitative and quantitative changes in microbial function under a given set of conditions. This is very important because of the potential variation in the characteristics of petroleum pollutants that might end up in the sediments as a result of an oil spill.

In our long-term effects study, the longest exposure time was 1.5 years in sediments exposed to 50 ppt fresh crude oil. Exposure to lower concentrations of crude oil for longer periods of time are required so that we can predict the expected length and severity of an impact soon after a spill. On the bottom of Kasitsna Bay, there is a set of treated sediments that, if analyzed, would provide much of that information. After 1.5 years exposure to 50 ppt fresh crude oil, there was some evidence that the initial effects were diminishing but the microbial functions were far from being normal.

C. There were two types of observations that we were not able to make during our Kasitsna Bay study which would be very important to documenting actual crude oil effects under spill conditions. Both of these studies could be conducted at the same time. One would include studying the effects of crude oil which had settled to the bottom via natural means rather than trying to stimulate these conditions by overlaying sediment with treated sediment. The other study would document the actual transfer of detrital carbon from the sediments into the food chain. Such a study could be conducted in large tanks and the analysis would be conducted by a multidisciplinary group of investigators which would include microbiologists, benthic ecologists, hydrocarbon and nutrient chemists and planktologists.

D. In addition to these studies, we recommend that a comprehensive plan be established for measuring the environmental impact of an actual oil spill. Studying such a spill is the best way to obtain actual effects data. The only way that this can be accomplished is to have a plan of sampling and experimental design prior to the spill.

CRUDE OIL WEATHERING

Section V.

I. Summary of objectives, conclusions, and implications with respect to OCS oil and gas development.

A. Objectives and background

The primary goal of this study was to examine the biological and chemical impact of fresh and weathered crude oil after its incorporation into sub-Arctic sedimentary regimes. Results of the hydrocarbon analyses from these experiments are presented in this section.

In an effort to assist Drs. Griffiths and Morita in this program, the Environmental Chemistry and Geochemistry Division of Science Applications, Inc. (SAI) undertook detailed chemical analyses of the sediment samples used in these experiments. Specifically, hydrocarbon profiles (concentrations) in control and experimental sedimentary plots were supplemented with three different levels of fresh and artificially weathered Cook Inlet crude oil and then examined: first, after the initial treatment, and second, after one year of natural weathering in the sedimentary regime at Kasitsna Bay, Alaska. Additional studies were also undertaken in Sadie Cove, Alaska, where oiled sediments were augmented with chitin and starch before deployment into the field, to determine if biotic weathering processes were controlled by limited nutrient concentrations.

B. Implications for off-shore oil and gas development

The results presented here support the conclusion that in a major oil spill event in the sub-Arctic marine environment, the most significant weathering of the oil will occur at the air/sea interface or in the water column before the oil is incorporated into the sedimentary regime. This is particularly true in fine-grain sediment matrices in low-energy environments. Once levels of fresh and weathered Cook Inlet crude oil reached concentrations in excess of 1 ppt in the sediment plots examined in the study, very little additional weathering or loss of higher molecular weight *aromatic* hydrocarbons occurred. In sediments supplemented with 50 ppt fresh or weathered crude oil, nearly complete inhibition of microbiological utilization or selective removal of *aliphatic* hydrocarbons was also observed, especially for those sediments supplemented with fresh crude. Recovery of biological activity and selective utilization of aliphatic hydrocarbons did occur in the samples treated with fresh and weathered crude at 1 ppt. In sediments supplemented with 0.1 ppt crude oil, there was little or no evidence of either aliphatic or aromatic petroleum hydrocarbon contamination after one year. At that time, the 0.1 ppt samples appeared to contain only the same biogenic hydrocarbons observed in the non-treated control sediment samples from Kasitsna Bay.

In the study plots which were supplemented with 50 ppt oil plus added nutrients (starch and chitin), there was no evidence of any enhanced biotic recovery or selective hydrocarbon utilization with either fresh or weathered crude oil. This suggests inhibition of biological processes from the high levels of toxic aromatic compounds in the oil itself rather than inhibition from limited nutrient concentrations. To more accurately address the role of added nutrients in oil degradation, detailed analyses should be completed at lower oil concentrations in the presence and absence of nutrients. Also, experiments to assess the role of dissolved oxygen, grain-size, the energy (tidal and wave) input to the sedimentary environment, total organic carbon content and other factors such as total biomass, could be considered in future studies.

From the results obtained on the fresh and weathered crude oils and the sediment samples examined in this program, it appears that the maximum amount of weathering and removal (dissolution and evaporation) of toxic components can be achieved if spill clean up and treatment efforts are designed to prolong the time that the oil remains on the water surface or suspended in the water column. This may suggest limited use of dispersants or detergents in certain spill situations, particularly if damage to coastal zones is not imminent. Containment and recovery of the residual higher molecular weight materials should take precedence over other strategies such as chemical dispersal which may result in higher subtidal sediment loadings.

II. Methods

Techniques for artificially weathering Cook Inlet crude oil and subsequent treatment, homogenization, and deployment of sediment into the experimental trays for in situ weathering are described in Section IV. Subsamples of the treated and control sediments from the experimental trays were collected and frozen at the initiation of the experiment and again after one year in the field. All frozen sediment samples were shipped on ice to SAI's Trace Environmental Chemistry Laboratory in one lot on 17 October 1980, where they were subsequently stored at -4°C until analyses were begun.

A. Extraction

Each sediment sample was extracted using a shakertable procedure which is similar to that described by Payne et al. (1978) and Brown et al. (1980) and which has been shown to yield comparable results to Soxhlet extraction (MacLeod and Fischer, 1980; and Payne et al., 1979). Briefly, the thawed sediment was placed in tared 500 ml Teflon jars and a wet weight was determined. Approximately 50 ml of methanol was added to the sediment for water removal, and the jars were sealed and agitated on a shaker table for 15 minutes. The jars were then centrifuged at 3000 rpm for 20 minutes at room temperature and the supernatant was decanted off and saved, and the drying procedure repeated. After the second drying step, 150 ml of methylene chloride (CH_2Cl_2) and methanol (65:35 v/v) were added to the jars and agitation was continued for 12 hours. The samples were centrifuged, the superantant saved, and the procedure was repeated with the agitation occurring for a period of 6 hours. The methanol-water washes and the methanol-methylene chloride extracts were combined in a separatory funnel and back extracted with 400-500 ml of saturated sodium chloride in distilled water which had been previously extracted with hexane. The lower layer (CH_2Cl_2) was removed and the water phase was back extracted with three additional 100 ml aliquots of CH_2Cl_2. The combined CH_2Cl_2 extracts were concentrated to approximately 100 ml using a Kuderna-Danish (K-D) apparatus, and dried by passage through a column of sodium sulfate followed by additional elution with CH_2Cl_2. The dried extract was concentrated to about 10 ml using a K-D apparatus and solvent exchanged (3x) into hexane, followed by solvent reduction to 1-2 ml in preparation for column chromatography.

B. Liquid column chromatography

To fractionate the sediment extracts, a three-part fractionation scheme was employed to separate the aliphatic, aromatic, and polar compounds (Payne, et al., 1980). A 10 mm I.D. x 23 cm long column with a 16 ml pore volume was packed with 1.5 cm of activated copper at the base of the column (to remove elemental sulfur), followed by a hexane slurry of 60/200-mesh silica gel that had been cleaned with CH_2Cl_2 and activated at 210°C for 24 hours. The elution scheme was as follows:

Fraction/Solvent	Amount	Compound Class
1. Hexane	30 ml	Aliphatic hydrocarbons
2. Hexane:Benzene 50:50	45 ml	Aromatic hydrocarbons
3. 50% CH_3OH in CH_2Cl_2	60 ml	Polar compounds

C. Gas chromatographic analysis

All gas chromatographic results were obtained on a Hewlett-Packard 5840A gas chromatograph equipped with an 18835A glass capillary inlet system and flame ionization detector. The microprocessor-based instrument was interfaced to a Texas Instruments Silent 700 ASR data terminal equipped with casette tape drive, allowing permanent storage of calibration data, retention times, and peak areas required for the data reduction system.

A 30-meter J&W Scientific Co. SE-54 wall-coated open tubular fused silica capillary column was utilized for the desired chromatographic separations. Temperature programming used with this column included:

Initial Temperature	50°C for 5 minutes
Program Rate	3.5°C/min
Final Temperature	275° for 60 minutes

The injection port and detector were maintained at 280° and 350°C, respectively. All injections were made in the splitless

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mode of operation with an injection port backflush 1 minute into the run.

Constant injection volumes of 1.0 μl were analyzed automatically using a Hewlett-Packard model 7671A Automatic Liquid Sampler, increasing precision substantially relative to manual injection.

D. Gas chromatogram data reduction

Hydrocarbon concentrations for individual resolved peaks in each gas chromatogram were calculated on a DEC-10 System Computer using the formula given in equation 1. This particular example is of the program used for seawater analysis. Operator-controlled modification of the DEC-10 program allows similar data reduction on sediments, tissues, or individual oil (mousse) samples.

$$\mu\text{g compound X/L seawater} = (A_x) \times (\text{R.F.}) \times \left[\frac{\text{P.I.V.} + 1}{\text{Inj.S.Vol.}} \; \frac{\text{Pre-C.S. Vol.}}{\text{Post-C.S.Vol.}} \; \frac{100}{\%\text{NSL on LC}} \times \frac{100}{\%\text{DW/FW}} \times \frac{1}{\text{liters}}\right] \quad (1)$$

where:

A_x = the area of peak X as integrated by the gas chromatograph (in arbitrary GC area units)

R.F. = the response factor (in units of μg/GC area unit)

P.I.V. + 1 = the post-injection volume (in μl) from which a 1-μl aliquot had been removed for analysis by GC (measured by syringe immediately following sample injection)

Inj.S.Vol. = the volume of sample injected into the GC (always 1.0 μl as measured by an HP Automatic Liquid Sampler)

Pre-C.S.Vol. & Post-C.S. Vol. = the total solvent volumes before and after an aliquot is removed for gravimetric analysis on a Cahn electrobalance

%NSL on LC = the percent of sample non-saponifiable lipid used for SiO_2 column chromatography

%DW/FW = the percent dry weight of wet weight in the sediment tissue, or oil sample being analyzed

liters = liters of seawater initially extracted (or grams wet weight of oil or sediment).

During analysis of the extracts, the 5840A gas chromatograph was recalibrated after every 8 to 10 injections, and individual response factors were calculated for all detected even and odd n-alkanes between nC_8 and nC_{32}. Concentrations of other components (e.g., branched and cyclic) that eluted between the major n-alkanes were calculated by linear interpolation of the adjacent n-alkane response factors and the unknown compound peak's KOVAT index. By incorporating the post-injection volume (PIV) into the calculation, the amount of hydrocarbons measured in the injected sample were converted to the total hydrocarbon concentration in the sample.

Unresolved complex mixtures (UCM's) were measured in triplicate by planimetry; the planimeter area was converted to the gas chromatograph's standard area units at a given attenuation and then quantitated using the average response factors of all the n-alkanes occurring within the range of the UCM, as shown in equation 2.

$$\frac{\mu g\ UCM}{liter} = Area_p \times (Conv.\ F) \times \frac{S.\ Att.}{Ref.\ Att.} \times (R.F._{a-b}) \times [\ldots] \quad (2)$$

where:

Area = UCM area in arbitrary planimeter units,

Conv. F. = a factor for converting arbitrary plainmeter units to GC area units at a specific GC attenuation, S.Att. and Ref. Att. = the GC attenuation at which the sample chromatogram was run and the reference attenuation to determine the conversion factor (Conv. F.), respectively

$R.F._{a-b}$ = the mean response factor for all sequential n-alkanes (with carbon numbers a to b) whose retention times fall within the retention time window of the UCM, and

[...] = the same parameters enclosed in brackets in equation 1.

Confirmation of KOVAT index assignment to n-alkanes was done by computer correlation with n-alkane standard retention times and direct data-reduction-operator input.

Assignment of KOVAT index to each branched or cyclic compound eluting between the n-alkanes was done by interpolation using the unknown compound and adjacent n-alkane retention times. Assignment of KOVAT indices to peaks in the aromatic fraction was made by direct correlation of unknown peaks with retention times from the n-alkane and aromatic standard runs completed prior to sample injection (Payne, et al., 1978b).

E. Capillary gas chromatography mass spectrometry

Selected extractable organic compounds previously analyzed by fused silica capillary column-FID GC were also subjected to fused silica capillary gas chromatography/mass spectrometry (GC/MS). A 30-meter J&W Scientific Co. SE-54 capillary column (0.25-mm I.D. with a film thickness of 25 μm was used to achieve chromatographic separation in a Finnigan 4021 quadrupole mass spectrometer. The capillary system was operated in the splitless (Grob-type) mode. The static time upon injection was 0.8 min, after which time the injection port was back-flushed with the split and septum sweep flows at a combined rate of 35 ml/min. Linear velocity was set at 35 cm/sec, which gave a flow rate of 1.18 ml/min. The GC was programmed to remain isothermal at 30°C for 1.5 min following injection, elevated at 4°C/min from 30 to 160°, and 8°C/min from 160-275°, after which the oven was held isothermally at 275°C for approximately 20 minutes.

The flexible fused silica column was routed directly into the ion source of the mass spectrometer, which was operated in the electron impact mode at 70eV with the lens potentials optimized for maximum ion transmission. The quadrupole offset and offset programs were adjusted to yield a fragmentation ratio for perfluorotributylamine m/e 69-to-219 of 4:1. This tuning yields quadrupole electron impact spectra that are comparable to magnetic sector electron impact spectra, thereby allowing optimal matches in the computer search routines used in the INOCS data system that scans the quadrupole rods from 35-475 amu in 0.95 sec. A hold time of 0.05 sec between scans allows the electronics to stabilize prior to the next scan. The mass spectrometer was tuned at the beginning of each day using perfluorotributylamine. A calibration was accomplished with a routine diagnostic fit of 2% mass accuracy. Prior to analysis of samples, standard mixtures of n-alkanes, pristane, phytane, and mixed aromatic hydrocarbons were injected.

III. Results

A. Time zero samples

Figure 66 presents the FID capillary gas chromatograms obtained on the control sediment samples taken from Kasitsna Bay at the beginning of the experiments. The most characteristic feature in

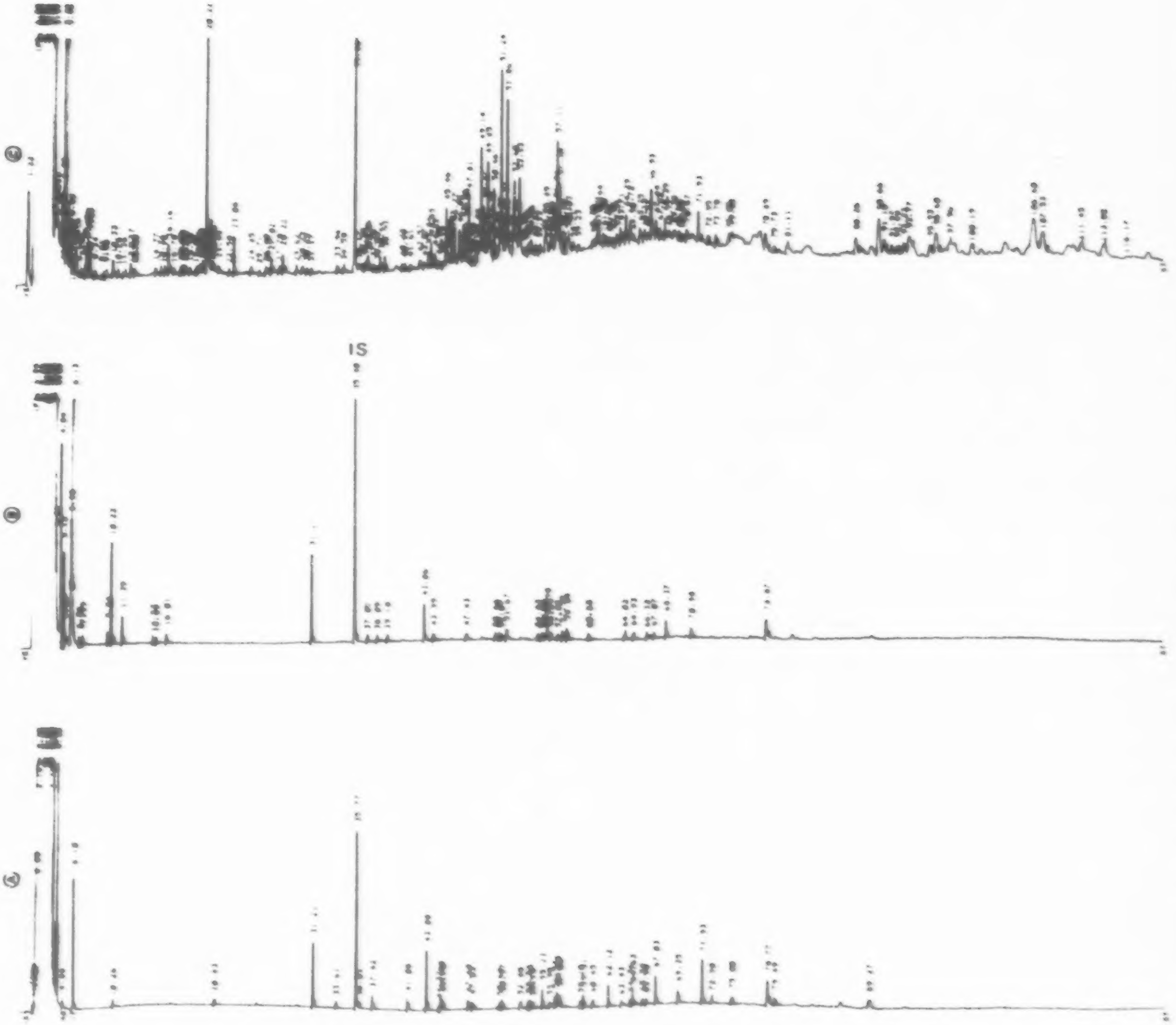

Figure 66. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction; B, the aromatic fraction and C, the polar fraction extracts obtained from time zero control sediment samples from Kasitsna Bay.

the aliphatic fraction chromatogram, A, is the predominance of odd numbered n-alkanes in the molecular weight range of nC_{21} to nC_{29}, (RT 56.53; 62.12; 67.03; 71.93; 78.77) reflecting biogenic input. The sample also contains very low levels of nC_{17} and pristane (RT 44.55; 44.75) and nC_{18} and phytane (RT 47.40; 47.83). The three major components at retention times 31.21, 35.77 and 43.08 are internal spikes and a GC recovery standard (triisopropylbenzene, 31.21; n-decylcyclohexane, 43.08; and hexamethylbenzene, 35.77, respectively). There is no apparent evidence of any petroleum contamination and there appears to be a small cluster of branched and unsaturated biogenic hydrocarbons between nC_{20} and nC_{21}. The aromatic fraction chromatogram, B, from this sample shows very little contamination of any kind, with only subnanogram per gram-dry-weight components present. The polar fraction chromatogram, C, does show evidence of several polar materials which are currently undergoing analysis by GC/MS. From GC/MS analyses of similar sediment samples, the identities of these peaks are suspected to be long chain fatty acid esters of biogenic origin.

Tables 57 and 58 present the reduced quantitative data obtained from the capillary FID gas chromatographic runs of these and all the other sediment samples analyzed as part of this program. The data in Tables 57 and 58 illustrate several interesting quantitative aspects which should be considered when interpreting the results. First, the background levels of hydrocarbons in the control samples from Kasitsna Bay at times zero and one year were both extremely low. In neither case was an Unresolved Complex Mixture (UCM) present, and the highest hydrocarbon concentration in these two samples was only six micrograms per gram dry weight. The odd to even n-alkane ratios for these samples (1 and 625) were high, ranging

Table 57. Reduced Aliphatic Hydrocarbon Data Derived from Flame Ionization Detector Capillary GC Analyses

Sediment Sample	OSU ID No	Time in Field (years)	Total Resolved ug/g	Total UCM* ug/g	Σ n-alkanes ug/g	Σ even n-alkanes ug/g	Σ odd n-alkanes ug/g	pristane + phytane / n-alkanes	odd alk / even alk	prist / nC_{17}	phy / nC_{18}	pri: / phyt	n-alk / branched
KASITSNA BAY CONTROL	1	0	5.89	0.	2.76	0.449	2.31	0.0157	5.15	0.758	0.	4--	0.883
KASITSNA BAY CONTROL	625	1	1.22	0.	0.533	0.06	0.473	0.	7.86	0.	0.	0.	0.776
FRESH CRUDE SPIKE 50ppt	5	0	2840.	4090.	1460.	742.	722.	0.0749	0.973	0.681	0.343	2.63	1.06
FRESH CRUDE SPIKE 1ppt	4	0	83.3	154.	52.8	24.2	28.6	0.055	1.18	0.630	0.411	2.18	0.690
FRESH CRUDE SPIKE 0.1ppt	3	0	11.3	12.8	5.03	2.11	2.92	0.596	1.39	0.601	0.380	2.40	0.803
WEATHERED CRUDE SPIKE 50	14	0	1530.	3020.	945.	460.	485.	0.104	1.06	0.721	0.326	2.75	1.62
WEATHERED CRUDE SPIKE 1	12	0	62.2	136.	40.3	19.3	21.0	0.0871	1.09	0.645	0.371	2.27	1.84
FRESH CRUDE SPIKE 50ppt	628	1R	2430.	1740.	1080.	560.	515.	0.0861	0.921	0.761	0.354	2.63	0.796
FRESH CRUDE SPIKE 50ppt	629	1R	2700.	3770.	1250.	645.	605.	0.0799	0.934	0.700	0.386	2.43	0.860
FRESH CRUDE SPIKE 50ppt	630	1R	2060.	3010.	950.	494.	458.	0.0832	0.927	0.708	0.357	2.41	0.862
FRESH CRUDE SPIKE 1ppt	631	1	21.3	98.1	10.9	4.89	6.04	0.144	1.24	1.28	0.709	2.40	1.06
FRESH CRUDE SPIKE 0.1ppt	634	1	3.29	13.1	1.09	0.179	0.912	0.0320	5.10	0.800	0.	3--	0.496
WEATHERED CRUDE SPIKE 50	637	1	1530.	3710.	949.	449.	499.	0.109	1.11	0.926	0.463	2.40	1.64
WEATHERED CRUDE SPIKE 1	640	1	14.3	319.	2.81	0.	2.81	0.	2--	0.	0.	0.	0.243
SADIE COVE CONTROL	206	0 -	26.1	0.	10.7	1.37	9.33	0.	6.81	0.	0.	0.	0.773
SADIE COVE OIL & STARCH	782	1	3760.	5200.	1605.	837.	768.	0.071	0.917	0.762	0.391	2.55	0.743
SADIE COVE OIL	779	1	4730.	6100.	1920.	983.	932.	0.0838	0.948	0.880	0.420	2.75	0.681
SADIE COVE OIL & CHITIN	780	1	4700.	6670.	1986.	1020.	968.	0.0630	0.951	0.626	0.345	2.14	0.732
COOK INLET CRUDE OIL													
FRESH	-	N.A.	84000	77600	33000	18300	14700	0.0703	0.804	0.673	0.331	2.43	0.648
WEATHERED	-	N.A.	38700.	54500.	24500.	11600.	12900.	0.0989	1.12	0.654	0.380	2.23	1.73

*UCM = Unresolved Complex Mixture

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Table 58. Reduced Aromatic Hydrocarbon Data Derived from Flame Ionization Detector Capillary GC Analyses.

Sediment Sample	OSU ID No	Time in Field (years)	Total Resolved ug/g	Total UCM* ug/g	Naphthalene (1185)**	2 Methyl-naphthalene (1295)	1 Methyl-naphthalene (1313)	Biphenyl (1381)	2,6 Dimethyl naphthalene (1404)	Fluorene (1586)	Phenan-threne (1786)	Anthracene	1 Methyl-phenanthrene	Fluoranthene	Pyrene
KASITSNA BAY CONTROL	1	0	3.08	0.	nd	nd	nd	nd	nd	nd	0.0262	nd	nd	nd	0.0392
KASITSNA BAY CONTROL	625	1	1.212	0.	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
FRESH CRUDE SPIKE 50ppt	5	0	1460.	1740.	58.0	137.	94.9	16.2	65.4	32.5	56.6	nd	18.0	nd	nd
FRESH CRUDE SPIKE 1ppt	4	0	12.2	22.3	0.363	0.960	0.570	0.0418	0.469	0.120	0.190	nd	0.0658	nd	nd
FRESH CRUDE SPIKE 0.1ppt	3	0	0.0289	0.0259	0.00025	0.00064	0.00037	nd	0.0032	nd	0.00032	nd	nd	0.0021	0.00012
WEATHERED CRUDE SPIKE 50	14	0	210.	824.	nd	5.59	4.77	nd	11.7	4.88	8.65	nd	nd	1.47	nd
WEATHERED CRUDE SPIKE 1	12	0	10.7	40.62	0.152	0.513	0.334	nd	0.467	0.150	0.255	nd	nd	nd	0.0376
FRESH CRUDE SPIKE 50ppt	628	1	469.	1235.	15.3	39.6	23.7	1.48	19.2	4.84	6.7	nd	2.48	1.00	nd
FRESH CRUDE SPIKE 50ppt	629	1	428.	880.	13.3	35.2	21.0	1.17	17.9	4.27	5.98	nd	2.40	0.840	nd
FRESH CRUDE SPIKE 50ppt	630	1	300.	2020.	9.75	30.4	18.6	nd	17.5	4.44	7.15	nd	2.95	nd	nd
FRESH CRUDE SPIKE 1ppt	631	1	9.33	29.7	0.163	0.517	0.433	0.0311	0.383	0.107	0.129	nd	0.0621	nd	nd
FRESH CRUDE SPIKE 0.1ppt	634	1	2.23	3.98	nd	0.0257	nd	nd	0.0321	nd	0.0307	nd	nd	nd	0.0185
WEATHERED CRUDE SPIKE 50	637	1	288.	901.	nd	5.19	4.07	nd	9.58	3.71	6.80	nd	nd	nd	nd
WEATHERED CRUDE SPIKE 1	640	1	12.28	51.84	0.0429	0.243	0.134	0.0234	0.420	0.195	0.137	nd	0.116	0.032	0.0297
SADIE COVE CONTROL	206	0	2.92	0	nd	nd	nd	nd	nd	nd	0.10258	nd	nd	0.02803	0.21073
SADIE COVE OIL & STARCH	782	1	781.	2010.	22.2	51.0	29.5	1.21	22.2	4.50	8.37	nd	2.64	nd	nd
SADIE COVE OIL	779	1	872.	2440.	32.6	75.0	43.2	nd	32.3	6.27	10.2	nd	5.71	2.04	nd
SADIE COVE OIL & CHITIN	780	1	1070.	1660.	33.8	77.8	45.7	1.80	31.0	6.24	9.99	nd	6.66	nd	nd
COOK INLET CRUDE OIL															
FRESH	-	N.A.	34700.	41600.	484.	1110.	644.	nd	540.	nd	190.	nd	nd	nd	nd
WEATHERED	-	N.A.	15400.	31400.	nd	nd	nd	nd	223.	102.	235.	nd	nd	nd	nd

* Unresolved complex mixture.
** Kovat indices in parentheses.
nd = not detected.

from 5.2 to 7.9, reflecting predominance of the odd n-alkanes of biogenic origin.

Figures 67 and 68 present gas chromatograms of the hexane and benzene fractions from the fresh Cook Inlet crude oil and the artificially weathered Cook Inlet crude oil (respectively) which were added to the sediment samples. Figure 67A clearly shows a high degree of complexity in the lower molecular weight range from nC_8 through nC_{12} although the aliphatic fraction is characterized, in general, by n-alkanes from nC_8 through nC_{32}. The aromatic fraction shows a number of lower molecular weight aromatic compounds in the range of KOVAT index 800 to KOVAT index 1500 (RT 10.09 to 35.79). These compounds were identified by GC/MS as alkyl substituted benzenes such as xylenes, ethylbenzene, trimethylbenzene and propylbenzenes. The large peak at RT 35.79 is the GC internal standard hexamethylbenzene. Also in this sample are peaks identified as napthalene (RT 24.64), 2-methylnaphthalene (RT 29.38), 1-methylnaphthalene (RT 30.11), 2,6-dimethylnapthalene (RT 33.87), and several low level alkyl substituted phenanthrenes, as shown by the data in Table 58.

Figure 68 shows the gas chromatograms of artificially weathered crude oil used to treat the sediment samples. The aliphatic fraction, Figure 68A, shows loss of the lower molecular weight n-alkanes below nC_{13}; however, the higher molecular weight materials are present at approximately the same ratios as in the starting crude oil. This is illustrated by the consistency in the pristane/phytane, pristane/ nC_{17}, and phytane/nC_{18} ratios for the fresh and weathered crude oils, as shown by the data in Table 57. The aromatic fraction of the artificially weathered crude shows nearly complete absence

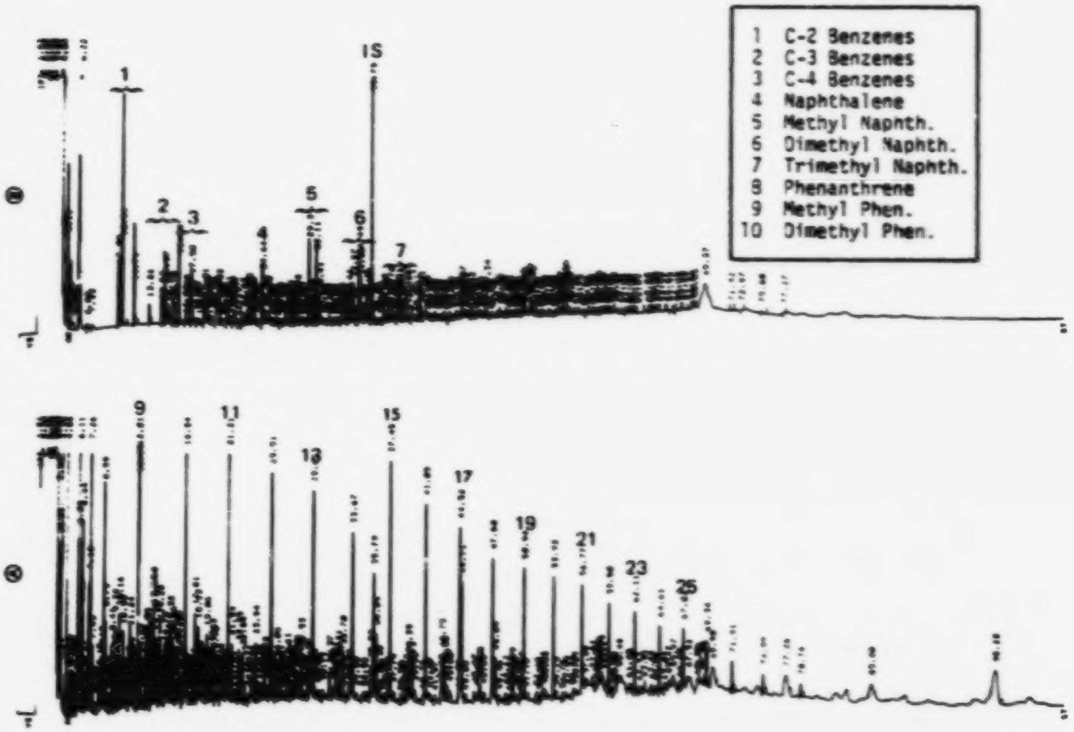

Figure 67. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction, and B, the aromatic fraction extracts obtained on the fresh Cook Inlet Crude Oil used to spike the Kasitsna Bay sediment samples.

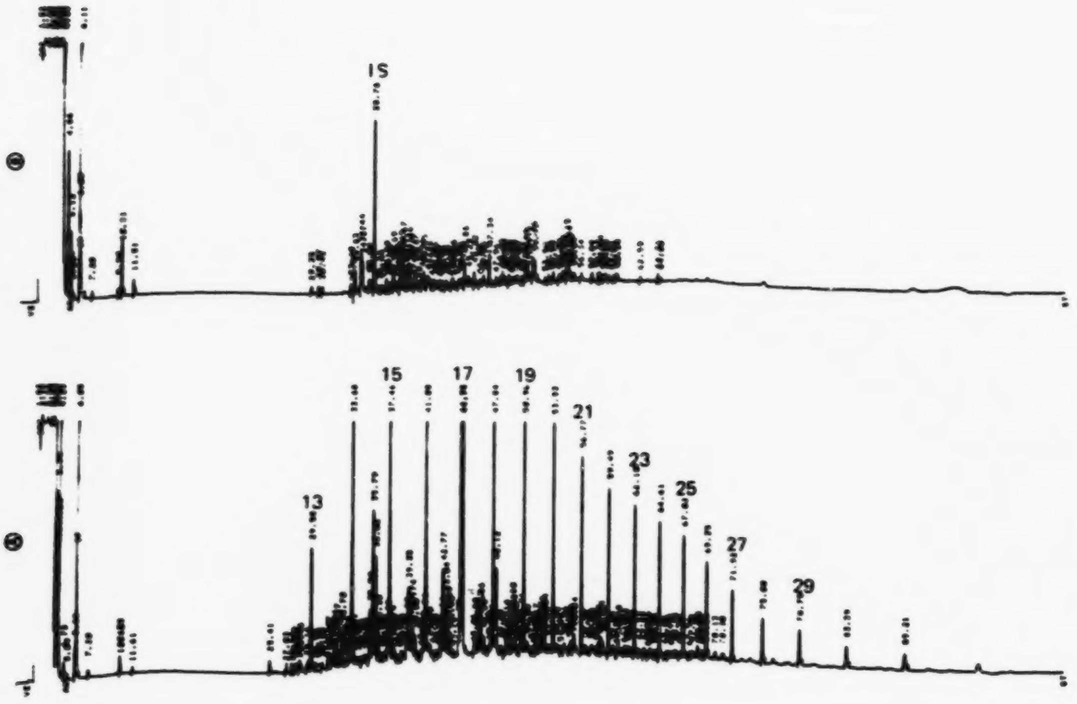

Figure 68. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction, and B, the aromatic fraction extracts obtained from the Artifically Weathered Cook Inlet Crude Oil used to spike the Kasitsna Bay sediment samples.

of the lower molecular weight hydrocarbons below dimethylnaphthalene; however, there still are several higher molecular weight polynuclears present. These are primarily phenanthrene at RT 47.34 (KOVAT 1790), 1-methylphenanthrene at RT 51.97 (KOVAT 1933), and fluoranthrene at RT 55.45 (KOVAT 2070). Higher molecular weight compounds such as benz(a)anthracene, benzo(e)pyrene, benzo(a)pyrene and perylene are not apparently present in either the starting or weathered Cook Inlet crude oil to an appreciable degree.

Figure 69 shows the gas chromatograms of the aliphatic, aromatic and polar fractions obtained on the Kasitsna Bay time zero sample treated with fresh crude oil at 1.0 ppt. The chromatograms obtained on the sediments treated with 50 ppt crude oil were essentially identical in appearance to those in Figure 69, and thus the 50 ppt sample's chromatograms are not shown here. Further, the concentrations of crude oil in the 50 ppt samples were at such a high level that only approximately 2% of the extractable materials could be effectively applied to the liquid chromatography columns for separation into aliphatic, aromatic and polar fractions. This allowed accurate quantitation of the materials but did not figuratively show the presence of the lower molecular weight compounds to the same degree as the 1.0 ppt samples where the entire sample could be fractionated and analyzed without prior dilution.

With regard to the chromatograms in Figure 69, the aliphatic fraction, A, is nearly identical to the aliphatic fraction of the starting fresh Cook Inlet crude oil shown in Figure 67. This is reflected qualitatively in the chromatograms presented in the figures and also quantitatively by the pristane/phytane, pristane/nC_{17}

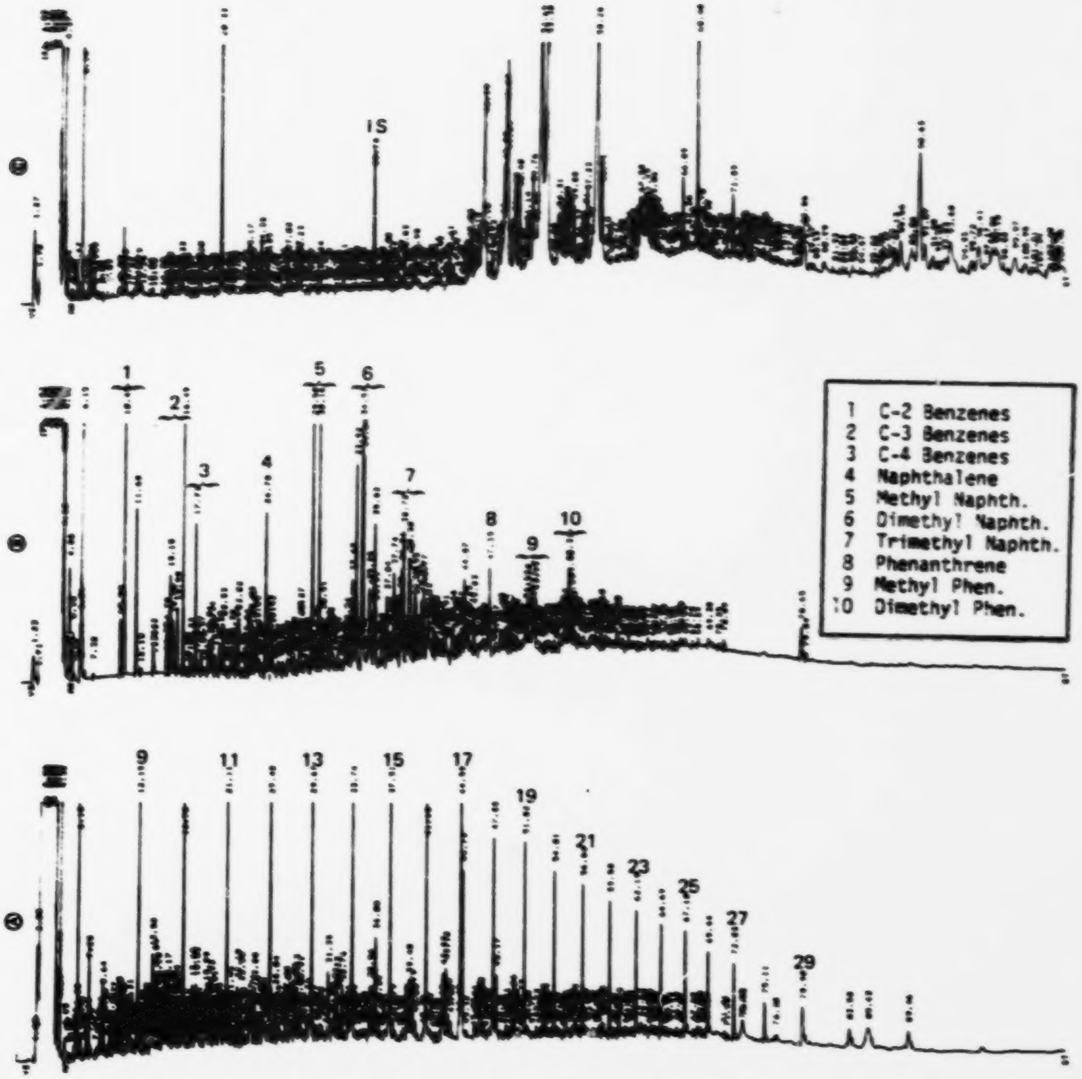

Figure 69. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction, B, the aromatic fraction, and C, the polar fraction extracts obtained from time zero Kasitsna Bay sediment samples which had been spiked with fresh Cook Inlet Crude Oil at 1 ppt.

and phytane/nC_{18} ratio data presented in Table 57. The suite of nC_{20}-nC_{21} branched/unsaturated compounds in the background control sample are completely masked in the treated sediment samples. The aromatic fractions of the treated sample show many of the same aromatic compounds in the naphthalene (KOVAT 1185) to pyrene (KOVAT 2124) range and the alkyl-substituted aromatic compounds at KOVAT indices 800 to 1012 as in the starting crude oil. The polar fraction of the oiled sediment at time zero shows many of the same biogenic compounds as in the Kasitsna Bay control sediment. This is particularly true of the compounds between retention times 46.99 and 68.40. These compounds are present at a greater apparent concentration in the oil treated sediment sample; however, examination of reduced chromatographic data output shows that this primarily reflects a smaller final sample extract volume resulting in more material being loaded on the fused silica capillary column.

Figure 70 presents the capillary chromatograms obtained on the time zero sediment samples treated with artificially weathered crude oil. The chromatograms are qualitatively very similar to those shown in Figure 68 which presented the weathered Cook Inlet crude used to treat the sediment samples. Aliphatics are virtually absent below nC_{13} as are aromatic compounds with KOVAT indices below 1300. A number of higher molecular weight polynuclear aromatic compounds can be identified in the weathered crude, and these are 2-methylnaphthalene at 29.38, 1-methylnaphthalene at 30.11, 2,6-dimethylnaphthalene at 33.88, fluorene at 40.61, phenanthrene at 47.41, 1-methylphenanthrene at 51.85 and fluoranthrene at 55.45. There appear to be no polynuclear aromatic hydrocarbons with molecular

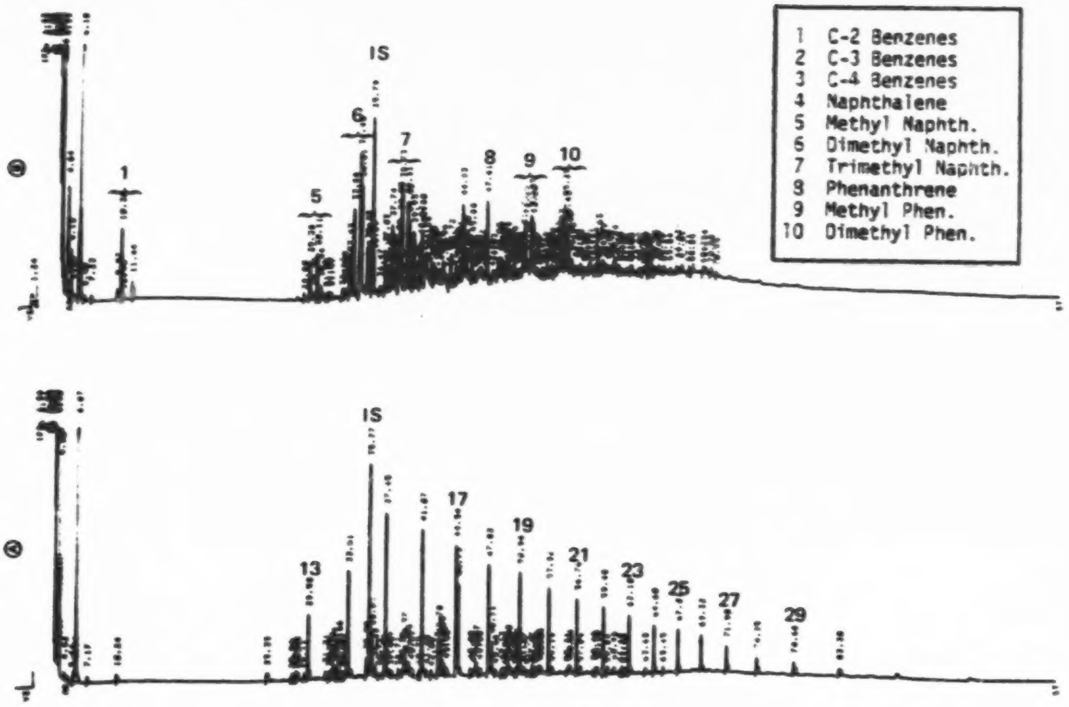

Figure 70. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction and B, the aromatic fraction extracts obtained from time zero Kasitsna Bay sediment samples spiked with Artifically Weathered Cook Inlet Crude Oil at 50 ppt.

weights greater than chrysene in the time zero artificially weathered sediment sample.

B. Time one year samples

After one year exposure, the trays were retrieved and subsamples of the sediments were collected. Figure 71 shows the chromatograms obtained on the aliphatic fraction of (A) the sediment treated with 50 ppt fresh crude oil at time zero, and (B, C, and D), the triplicate samples examined after one year of natural weathering. Several features are significant in this figure. The first and most obvious feature is the lack of any appreciable oil weathering at this high concentration level. This is reflected in the qualitative appearance of the chromatograms and in the data presented in Table 57. Specifically, the lower molecular weight n-alkanes from nC_8 through nC_{12}, and the branched and cyclic compounds occurring between KOVAT index 900 and 1000 appear to be nearly identical in all four samples.

Figure 72 graphically presents the concentration abundance of the n-alkanes in the sediment sample treated with 50 ppt fresh crude oil at time zero and again after one year of weathering in Kasitsna Bay. Note that in addition to the concentrations of the time zero and one year samples being very similar, the overall trends showing decreases in the higher molecular weight compounds are nearly identical for both samples, illustrating the lack of any appreciable selective weathering.

The similarity of the pristane/nC_{17} and phytane/nC_{18} ratios, as observed qualitatively in Figure 71 and Table 57, also illustrates the lack of any appreciable biotic or abiotic weathering in these

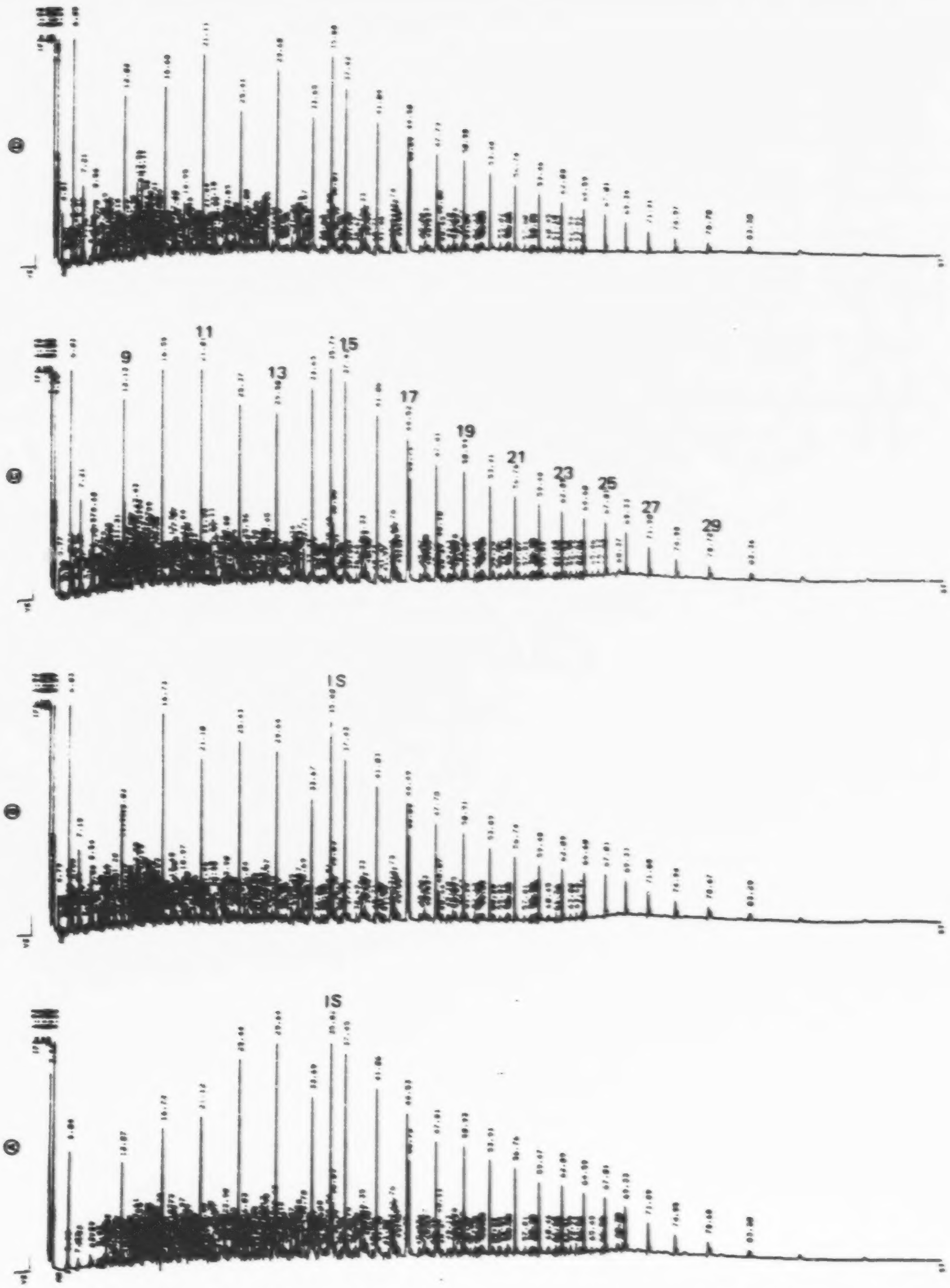

Figure 71. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction of the 50 ppt fresh Cook Inlet Crude Oil spiked into the sediment at time zero, and B, C, and D, the aliphatic fractions of the triplicate samples examined after one year of natural weathering in Kasitsna Bay.

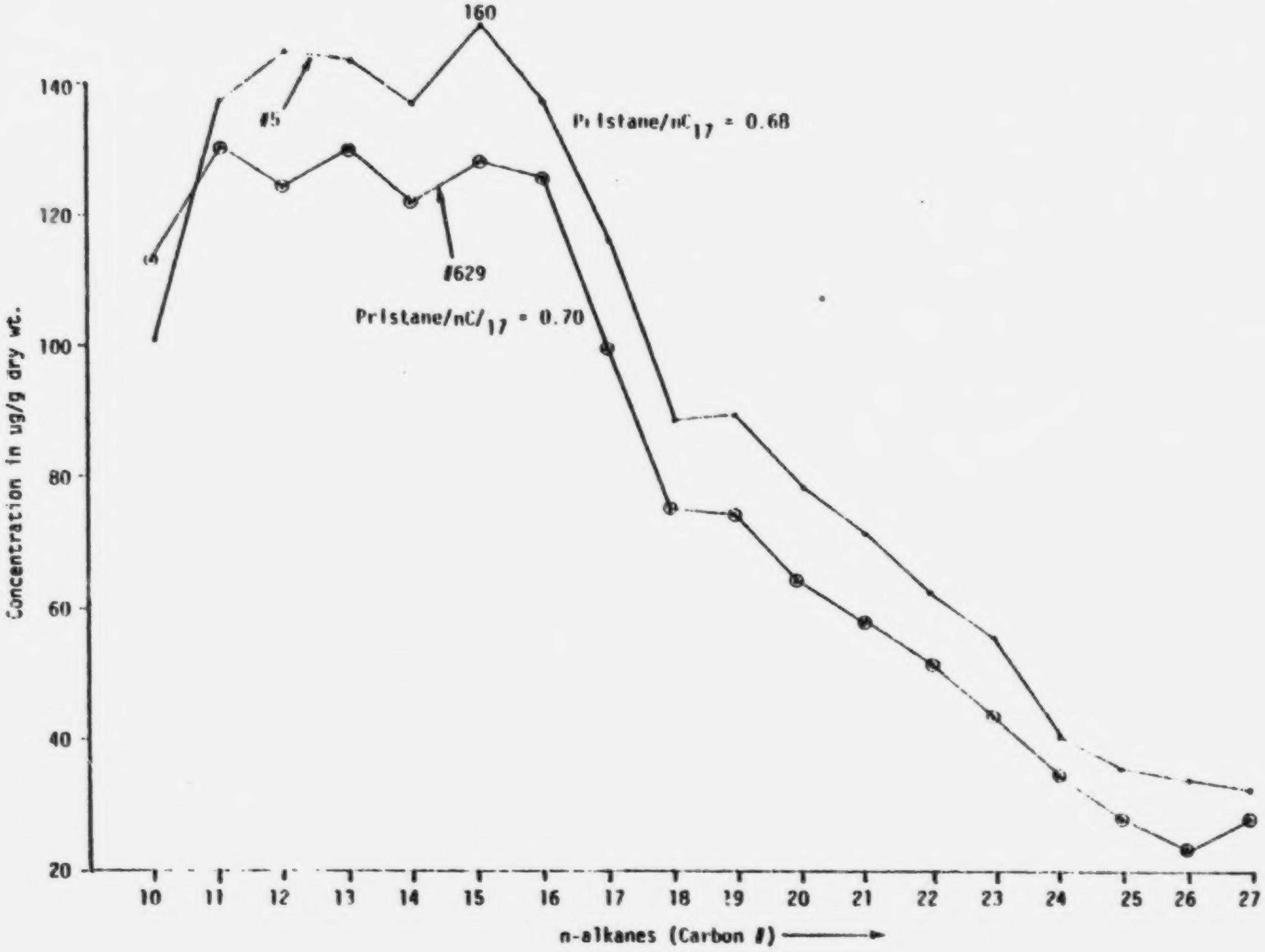

Figure 72. Concentration abundance of the n-alkanes in a sediment sample spiked with 50 parts per thousand fresh crude illustrating the time zero sample (·) and sample after one year of weathering (⊙).

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samples. The chromatographic profiles are essentially superimposable, reflecting the homogeniety of the initial treated sediment, the replicability of the weathering process in the field and the precision of the analytical method. Individual values for these three fractions are presented in Table 57, and the agreement of such features as the total n-alkanes, sum of the odd n-alkanes, even n-alkanes, pristane/phytane ratios, etc., is worthy of consideration.

Figure 73 presents the gas chromatograms of the aromatic fractions obtained on the sediment treated with 50 ppt fresh Cook Inlet at time zero (A) and the replicate fractions (B, C and D) obtained from analyses of the triplicate sediment samples after one year of natural weathering. As in Figure 71, there does not appear to be any selective weathering of the individual components present; however, examination of the reduced data in Table 58 and Figure 74 shows that some decreases in aromatic hydrocarbon concentrations did occur after 1 year. The apparent lower levels of material in chromatogram A (Figure 73) only reflect a larger final sample extract volume from which an aliquot was removed for analysis by GC. Figure 74A presents a graphical representation of the concentrations of eight selected aromatic compounds in the fresh 50 ppt treated sediments at time zero and time one year. While time zero levels of individual aromatic compounds ranged from 50 to 137 micrograms per gram dry weight (for napthalene through 2,6-dimethylnaphthalene), after one year these compounds were present at concentrations ranging from 15 to 40 micrograms per gram dry weight. The decreases in aromatic compounds from Kovat indices 1100 to 1500 were greater than the decreases in aromatics with Kovat indices ranging from 1500 to 2000. This presumably reflects two things: 1) the greater

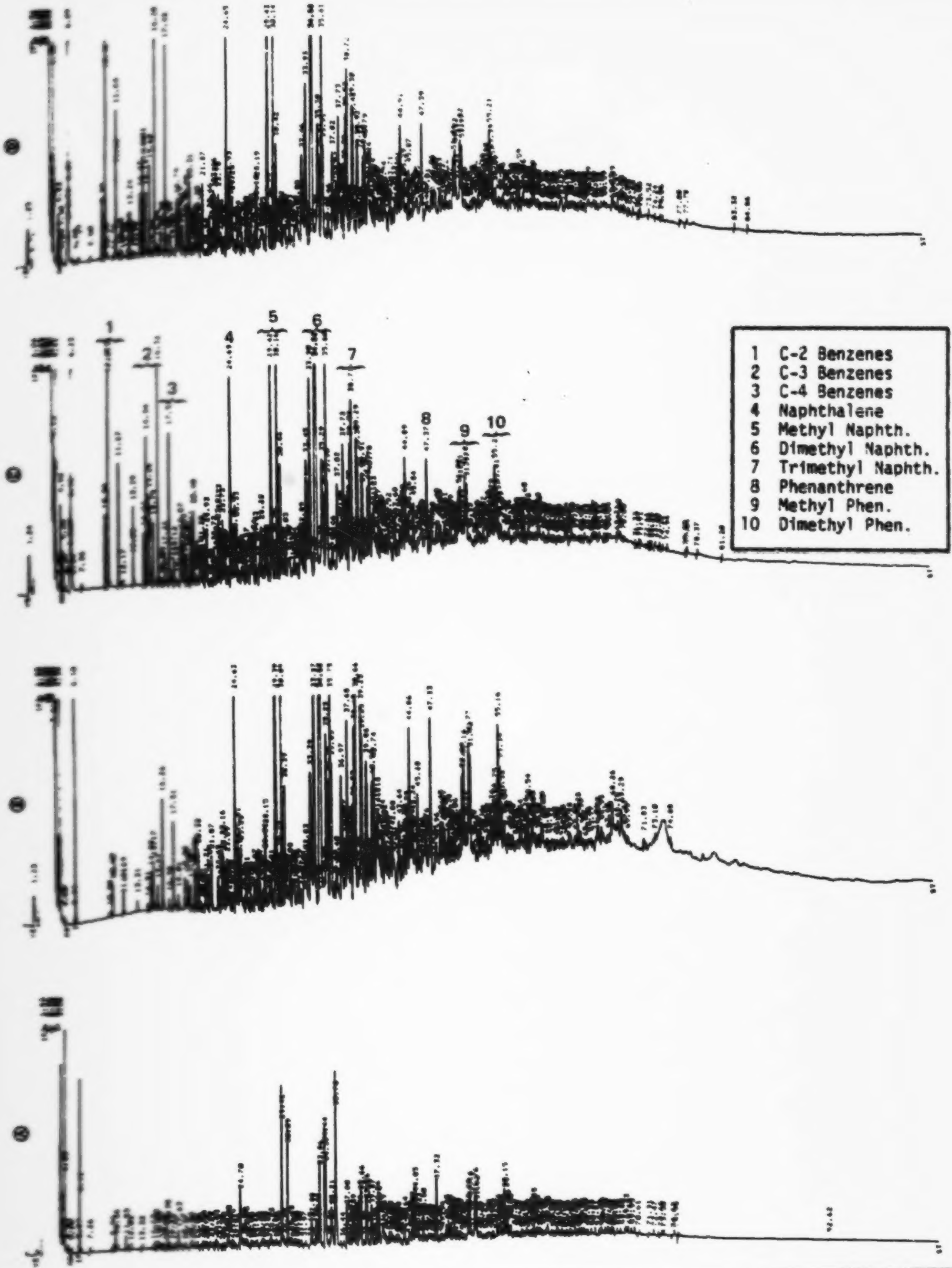

Figure 73. Flame Ionization Detector gas chromatograms of: A, the aromatic fraction of the 50 ppt fresh Cook Inlet Crude Oil spiked into the sediment at time zero and B, C, and D, the aromatic fractions of the triplicate samples examined after one year of natural weathering in Kasitsna Bay.

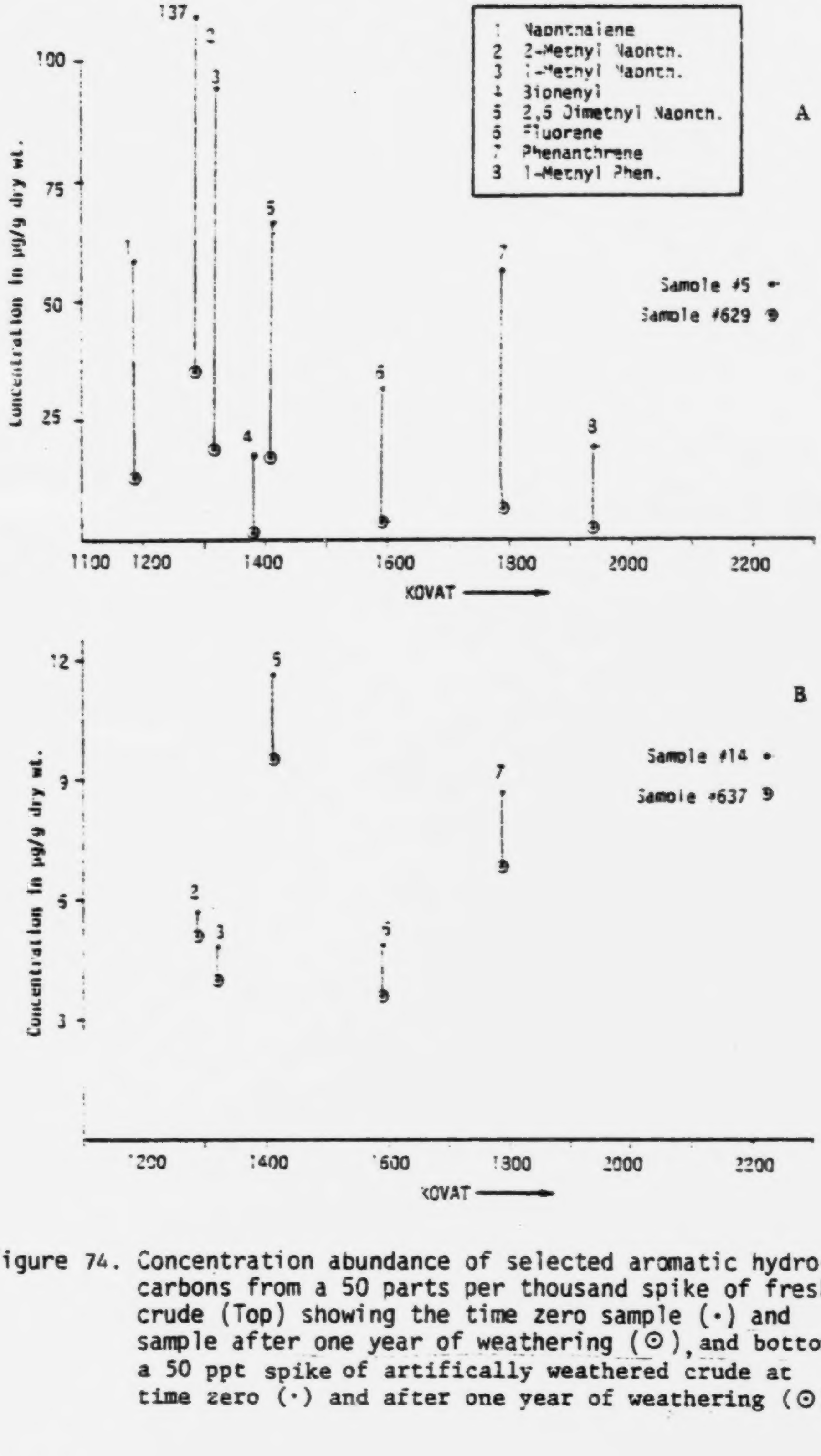

Figure 74. Concentration abundance of selected aromatic hydrocarbons from a 50 parts per thousand spike of fresh crude (Top) showing the time zero sample (·) and sample after one year of weathering (⊙), and bottom. a 50 ppt spike of artifically weathered crude at time zero (·) and after one year of weathering (⊙).

volatility and water solubility of the lower molecular weight aromatic compounds, and 2) the lower relative abundance of the higher molecular weight aromatics in the crude oil to begin with.

Figure 74B shows the relative losses of aromatic hydrocarbons in the artificially weathered crude oil added into the Kasitsna Bay sediments at time zero and time one year. This figure illustrates that much smaller relative changes occurred over the one year period. That is, the starting concentrations of aromatic compounds such as 2-methylnaphthalene through phenanthrene ranged between only 6 and 12 micrograms per gram dry weight of sediment when artificially weathered crude was used to treat the sample at time zero. These levels were not significantly reduced after one year of weathering in the sediments of Kasitsna Bay: the most significant weathering occurred while the oil was "artificially weathered" on the surface of a salt water aquarium before the oil was spiked into the sediment. Nevertheless, once these compounds are introduced into the sediments, they are not as rapidly removed as they would be from simple dissolution in the starting oil itself.

Figure 75 presents the gas chromatograms of the aliphatic and aromatic fractions obtained on the sediment treated with 1 ppt fresh-crude oil after one year of weathering in Kasitsna Bay. In comparison with Figure 69 which shows the starting 1.0 ppm material, it is clear that signficant weathering of the sample has occurred. This is reflected first in the significantly greater relative loss of the lower molecular weight alkanes below nC_{13}, presumably due to a combination of biological and abiotic (dissolution) processes.

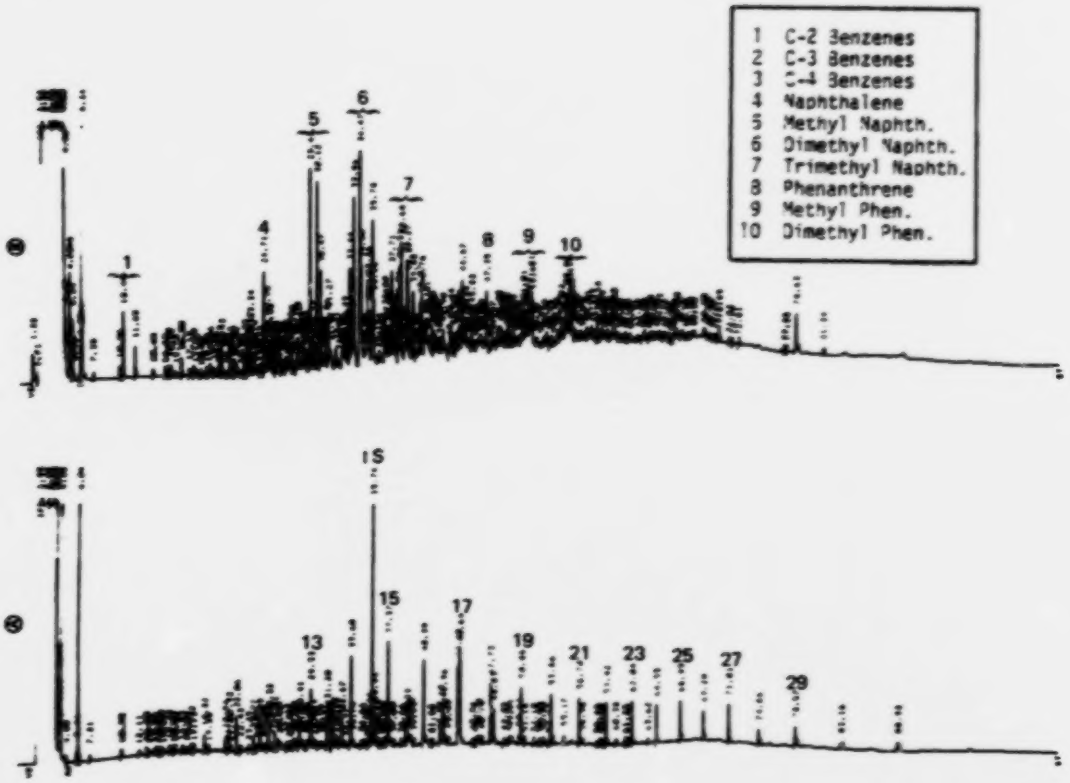

Figure 75. Flame Ionization Detector gas chromatograms of: A, the aliphatic fraction and B, the aromatic fraction extracts obtained from 1 ppt fresh Cook Inlet Crude Oil spiked sediments after one year of weathering in Kasitsna Bay.

Evidence of bio-chemical degradation is shown in examining the pristine/nC_{17} and phytane/nC_{18} levels in the aliphatic fraction in Figure 75 compared to the aliphatic fraction in Figure 69, and by examining the numerical values for these ratios in Table 57. The straight chain alkanes have been preferentially removed relative to the branched chain isoprenoids. The overall levels of other aliphatic hydrocarbons are also significantly reduced as illustrated qualitatively in Figure 75 and by the data in Table 57. Figure 76 graphically presents the concentration abundance of n-alkanes in the 1.0 ppt fresh crude oil sediment treated at time zero and after one year of natural weathering. All of the lower molecular weight alkanes below nC_{18} are significantly reduced by a factor of from 2 to 5 and the higher molecular weight n-alkanes are reduced by at least a factor of 2 compared to the sample taken at time zero. For the 1 ppt sample the total resolved hydrocarbons decreased from 83 to 21 µg/g dry weight during the year of exposure and the unresolved complex mixture decreased from 154 to 98 µg/g dry weight.

The aromatic fraction data in Figure 75B show somewhat less degradation compared to the aliphatic fraction. Compounds with molecular weights less than naphthalene (KOVAT <1185) are obviously removed due to a combination of biological and abiotic factors (dissolution and evaporation); however, compounds with molecular weights greater than 1-methylnaphthalene (KOVAT >1315) appear to be present in relatively identical concentrations compared to the starting materials. That is, while overall levels are slightly reduced as illustrated by the data in Table 58, the relative concentrations of the individual polynuclear aromatics are very similar

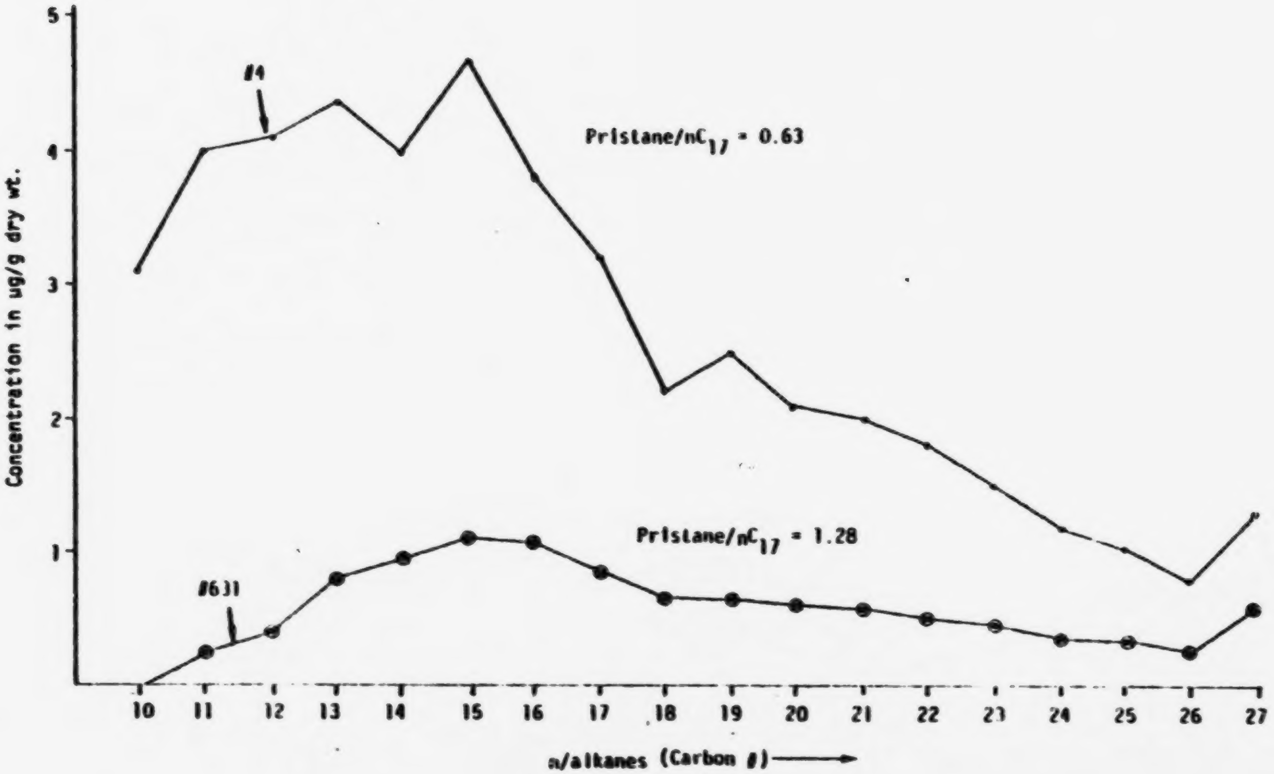

Figure 76. Concentration abundance of the n-alkanes for a sediment oil spike of 1.0 part per thousand fresh crude illustrating the time zero sample (·) and sample after one year of weathering (⊙).

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in the time zero and time one year samples. This is also reflected quite obviously by the qualitative appearance of the aromatic fractions shown in Figures 69B and 75B, respectively, and by the data presented in Figure 77A. Figure 77A graphically presents the relative changes in abundance of selected aromatic hydrocarbons from the sediment treated with 1 ppt fresh crude oil from time zero to one year. While the relative range of concentrations of all of the compounds in the time zero and one year samples are lower compared to the 50 ppt sample shown in Figure 74A, the overall concentrations of the time zero and naturally weathered 1 ppt samples are still relatively similar. This is particularly true of the higher molecular weight compounds, bi-phenyl, fluorene, phenanthrene and 1-methylphenanthrene. As in Figure 74A and B, the relative concentrations of artificially weathered aromatic compounds from the 1.0 ppt sample illustrated in 77B show that concentrations are in the same range in the artificially weathered sample as in the fresh sample after it had been weathered for a full year.

Clearly, while biological degradation of the aliphatic hydrocarbons (primarily n-alkane) occurred at the 1 ppt level, concommitant degradation of the higher molecular weight polynuclear aromatics compounds with molecular weights above that of methylnaphthalene did not occur at a significant level.

This lack of degradation of higher molecular weight PNA's at the 1.0 ppt level is also illustrated in Figure 78, which presents the aromatic fraction chromatograms of: A) the 1 ppt fresh crude added into the sediment at time zero; B) the aromatic fraction obtained from the 1 ppt sediment after one year of in situ weathering in Kasitsna Bay; and C) the aromatic fraction of the 1 ppt sediment

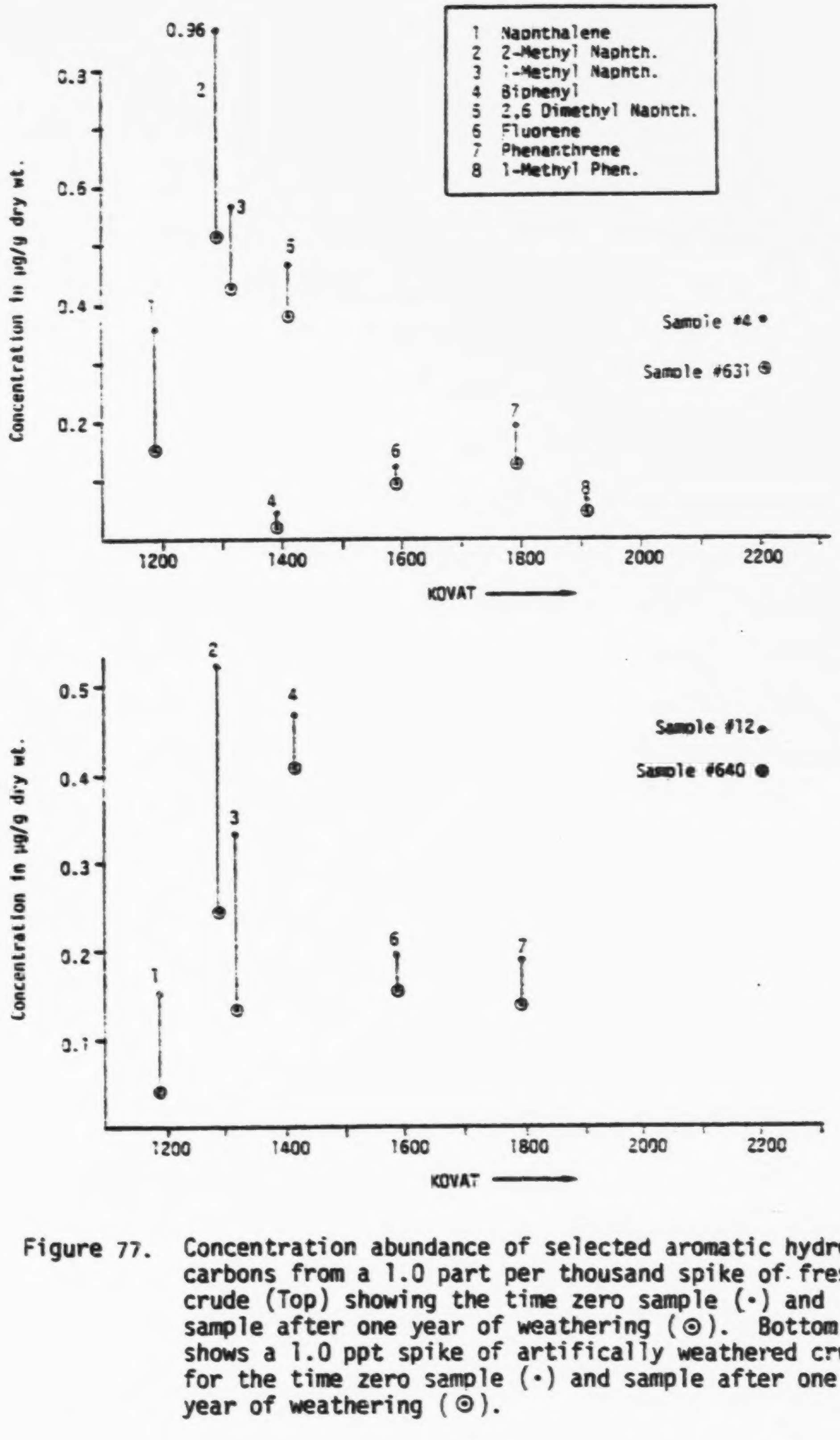

Figure 77. Concentration abundance of selected aromatic hydrocarbons from a 1.0 part per thousand spike of fresh crude (Top) showing the time zero sample (•) and sample after one year of weathering (⊙). Bottom shows a 1.0 ppt spike of artifically weathered crude for the time zero sample (•) and sample after one year of weathering (⊙).

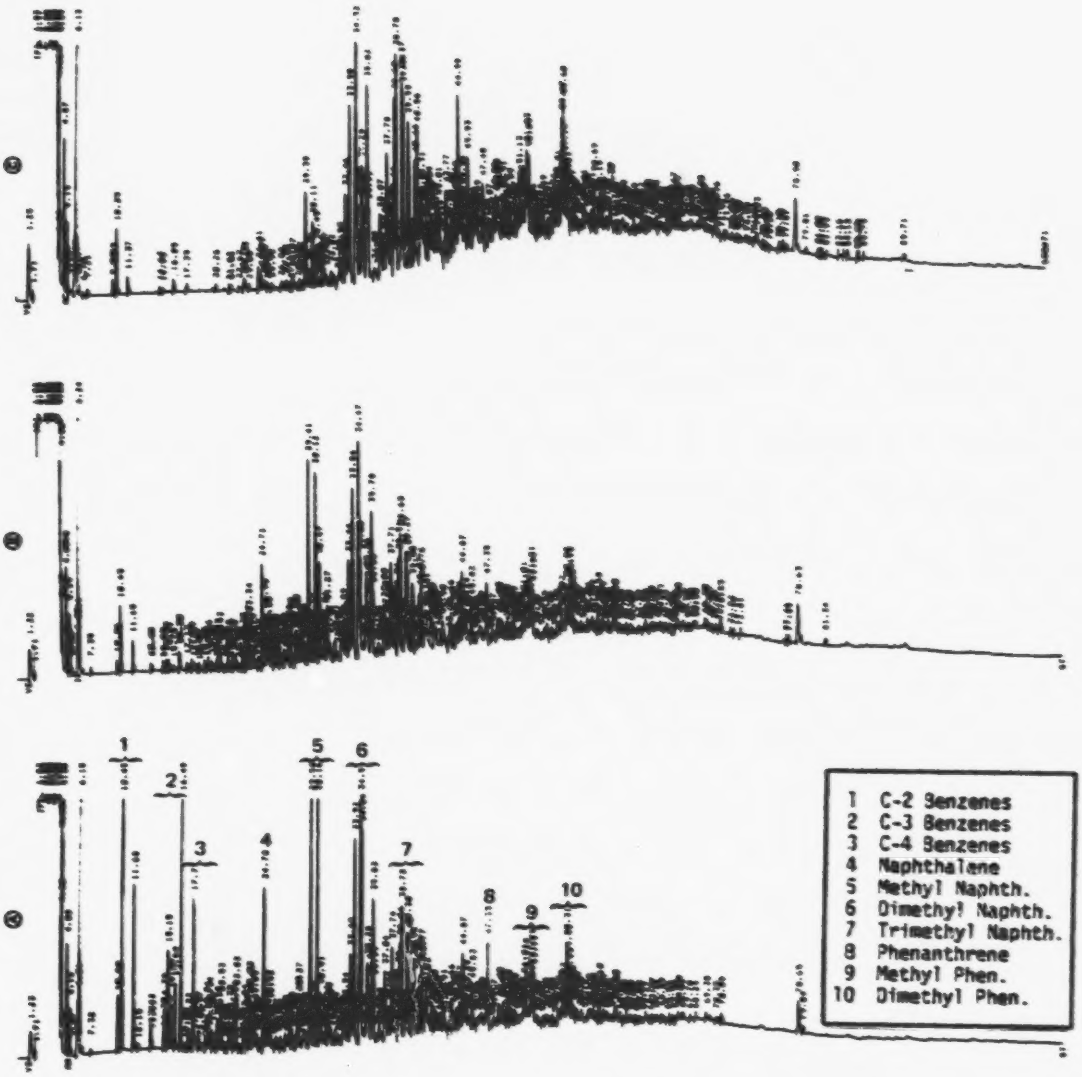

Figure 78. Flame Ionization Detector gas chromatograms of extracts of the aromatic fractions obtained from: A, the sediment spiked with 1 ppt fresh crude oil at time zero; B, the 1 ppt fresh crude sample after one year of natural weathering in Kasitsna Bay and C, the 1 ppt sediment sample spiked with artifically weathered crude oil after one year of additional weathering in Kasitsna Bay.

treated with artificially weathered crude oil after one year of additional weathering in Kasitsna Bay. Examination of chromatograms 78A and B shows that some loss of the lower molecular weight alkyl substituted benzenes at retention times 10.45, 11.68, 15.15, 15.90, 16.49 and 17.71 has occurred due to either evaporation or dissolution. Compounds with molecular weights greater than that of 1-methylnaphthalene at retention time 29.41 (B) are present in nearly identical relative concentrations. The chromatogram in 78C shows that the same compounds were also present in the "artificially weathered" oil which was added into the sediment after an additional year of natural weathering. This suggests that although many lower molecular weight aromatic compounds are removed from natural weathering of spilled oil while the oil is still at the surface, once the less water soluble and volatile higher molecular weight PNA's are incorporated into the sediment, additional degradative processes are extremely slow. Thus, while the relatively non-toxic aliphatic hydrocarbons are significantly degraded by biological processes in the sediments at 1 ppt, the more toxic aromatic compounds appear to be longer lived when introduced to the sediment from either fresh or weathered crude oil at similar levels.

Figure 79A presents the aliphatic fraction chromatogram obtained on the 0.1 ppt fresh crude oil added into the sediment at time zero. Figure 79 B, C and D presents the chromatograms of the same sediment after weathering in Kasitsna Bay for one year. Virtually all of the n-alkanes in the starting oil are no longer present in the sediment after one year of weathering. In fact, the only compounds of any significance in the aliphatic fraction of the fully weathered sediment are higher molecular weight odd n-alkanes,

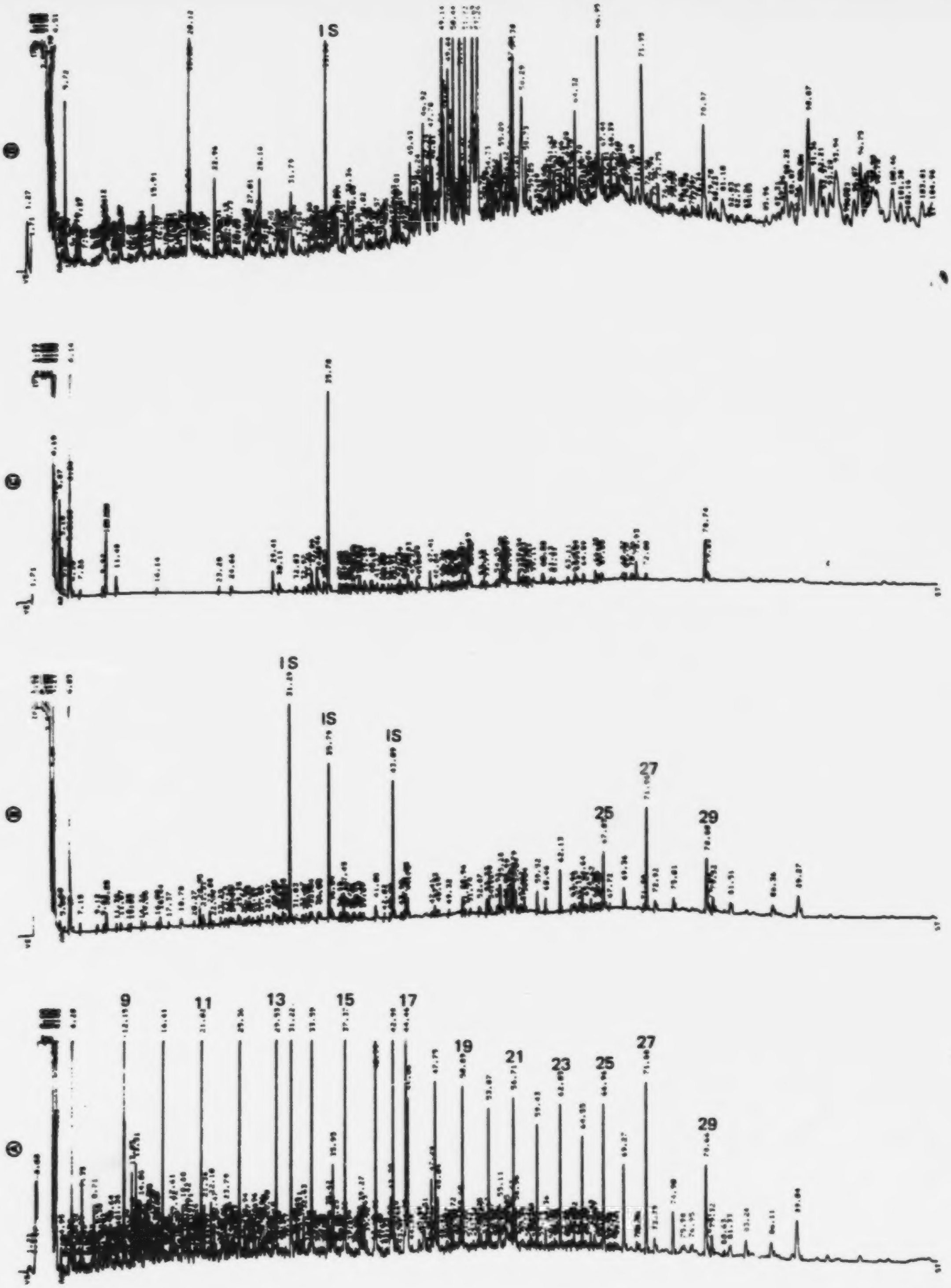

Figure 79. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction of the sediment spiked with 0.1 ppt Fresh Crude Oil at time zero and B, the aliphatic fraction, C, the aromatic fraction, and D, the polar fraction extracts obtained on the 0.1 ppt Fresh Crude Oil spiked sample after one year of Natural Weathering in Kasitsna Bay.

nC_{23}, nC_{25}, nC_{27}, and nC_{29}. These same compounds are also predominant in the fresh crude sample shown in Figure 79A. That is, instead of seeing a regular decrease in higher molecular weight n-alkanes from nC_{22} through nC_{32}, the odd carbons at 23, 25 and 27 from biogenic input are clearly present. These are the only compounds which remain in the sediment after one year, although there is some evidence that several unsaturated compounds between KOVAT indices 1900 and 2200 are present in Figure 79B. The aromatic fraction 79C shows only extremely low levels of residual materials with some evidence of pyrene perhaps remaining in the sediment at retention time 78.70. This compound was not detected in the starting oil to an appreciable degree; thus, its presence may reflect input from some other source.

Figure 80 shows the chromatograms of the aliphatic and aromatic fractions of the 50 ppt artificially weathered crude oil treated sediment one year of additional degradation in the sediment plots in Kasitsna Bay. Comparison of the sediments treated with the weathered crude oil at time zero, as shown in Figure 70, shows little or no change in the oil composition after one year of additional weathering. This is perhaps better illustrated in Figure 81, which presents the concentration abundance of the n-alkanes in the sediment treated with 50 ppt artificially weathered crude oil in the time zero sample and after one year of additional natural weathering. The data illustrate that all compounds below the level of nC_{14} are drastically reduced in both the starting material and the residual oil isolated after one year of natural weathering; however, the higher molecular weight compounds are not significantly altered.

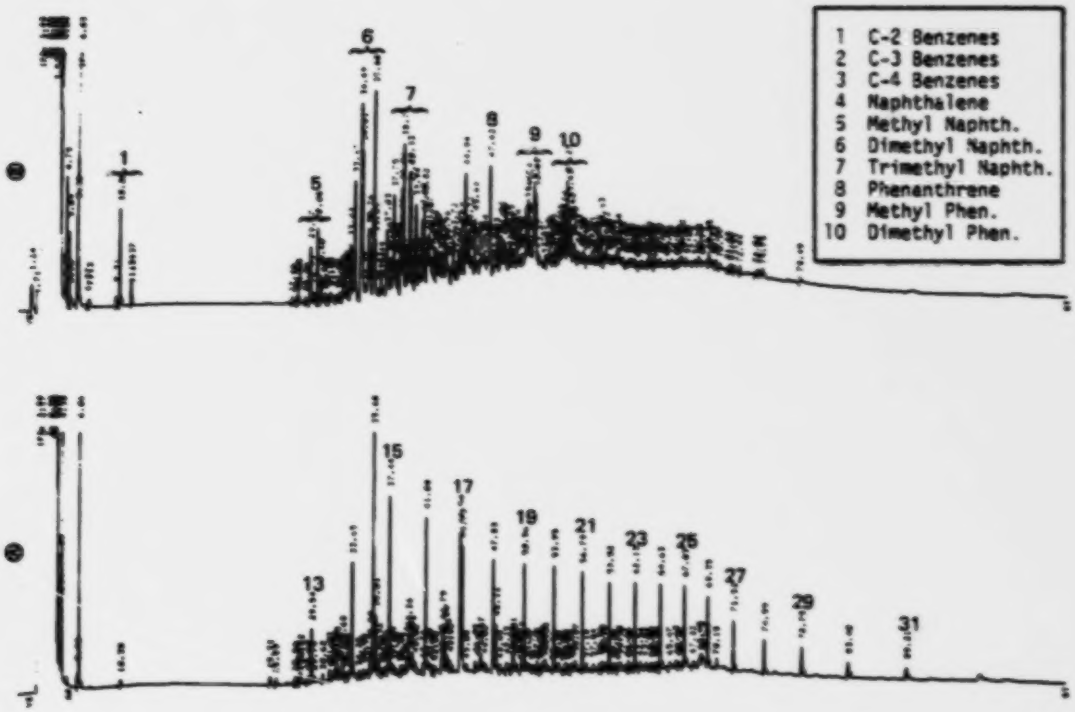

Figure 80. Flame Ionization Detector capillary gas chromatograms of: A, the aliphatic fraction and B, the Aromatic fraction extracts obtained from the time one year Kasitsna Bay sample spiked with Artifically Weathered Cook Inlet Crude Oil at 50 ppt.

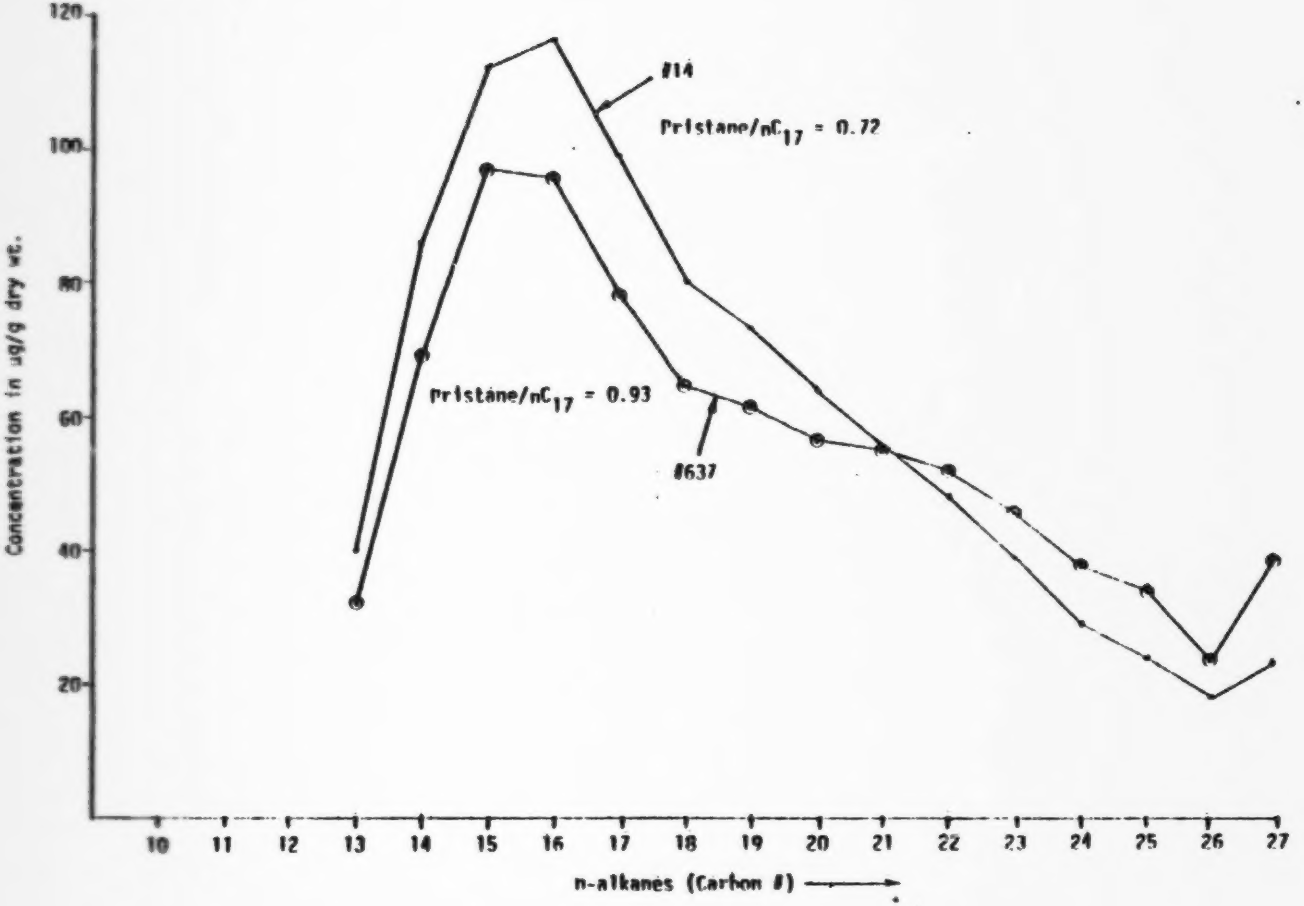

Figure 81. Concentration abundance of the n-alkanes for a sediment oil spike of 50 parts per thousand artifically weathered crude illustrating the time zero sample (·) and sample after one year of weathering (⊙).

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The corresponding data for the aromatic fraction of the 50 ppt artificially weathered crude are shown in Figure 74B. These data show that while the overall concentrations of the lower molecular weight mono and di-cylcic aromatic compounds were reduced in the weathered crude oil compared to the fresh crude oil, once the artificially weathered oil reached the sediment, further degradation and loss of the aromatic compounds did not occur.

When 1.0 ppt weathered crude oil was added into the sediments, much greater degradation and loss of the lower molecular weight n-alkanes occurred as illustrated by the data in Figure 82. In Figure 82, the loss of lower molecular weight aliphatic compounds can clearly be observed in the artificially weathered oil when it was added into the sediments. The sample collected after one year of weathering at Kasitsna Bay contained essentially no aliphatic hydrocarbons below nC_{24}. This was very similar to the case when 1.0 ppt fresh crude oil was added into the sediments and similar decreases in the aliphatic fraction were observed. The data in Figure 77B, however, show that the relative concentrations of aromatics in the 1.0 ppt weathered crude did not decrease significantly over the year period after the oil was introduced into the sediment. Quite clearly from these results, after fresh or weathered oil is incorporated into the sub-Arctic sedimentary regime at concentrations greater than 1.0 ppt, only limited additional degradation of the aromatic fraction occurs in periods up to one year.

C. Sadie cove oil/nutrient amended sediment experiments

Figure 83 presents the aliphatic fraction chromatograms obtained on (a) the 50 ppt oil plus starch, (b) 50 ppt oil alone and

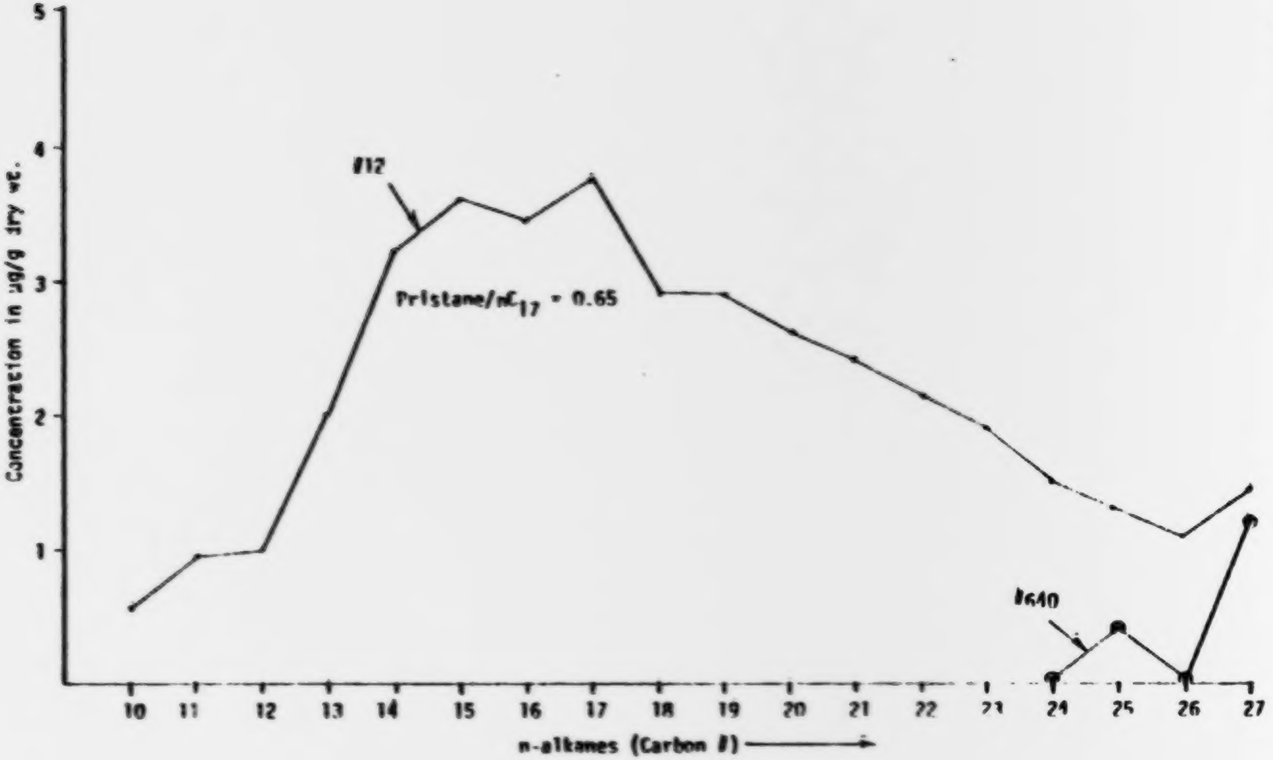

Figure 82. Concentration abundance of the n-alkanes for a sediment oil spike of 1.0 parts per thousand artifically weathered crude illustrating the time zero sample (·) and sample after one year of weathering (⊙).

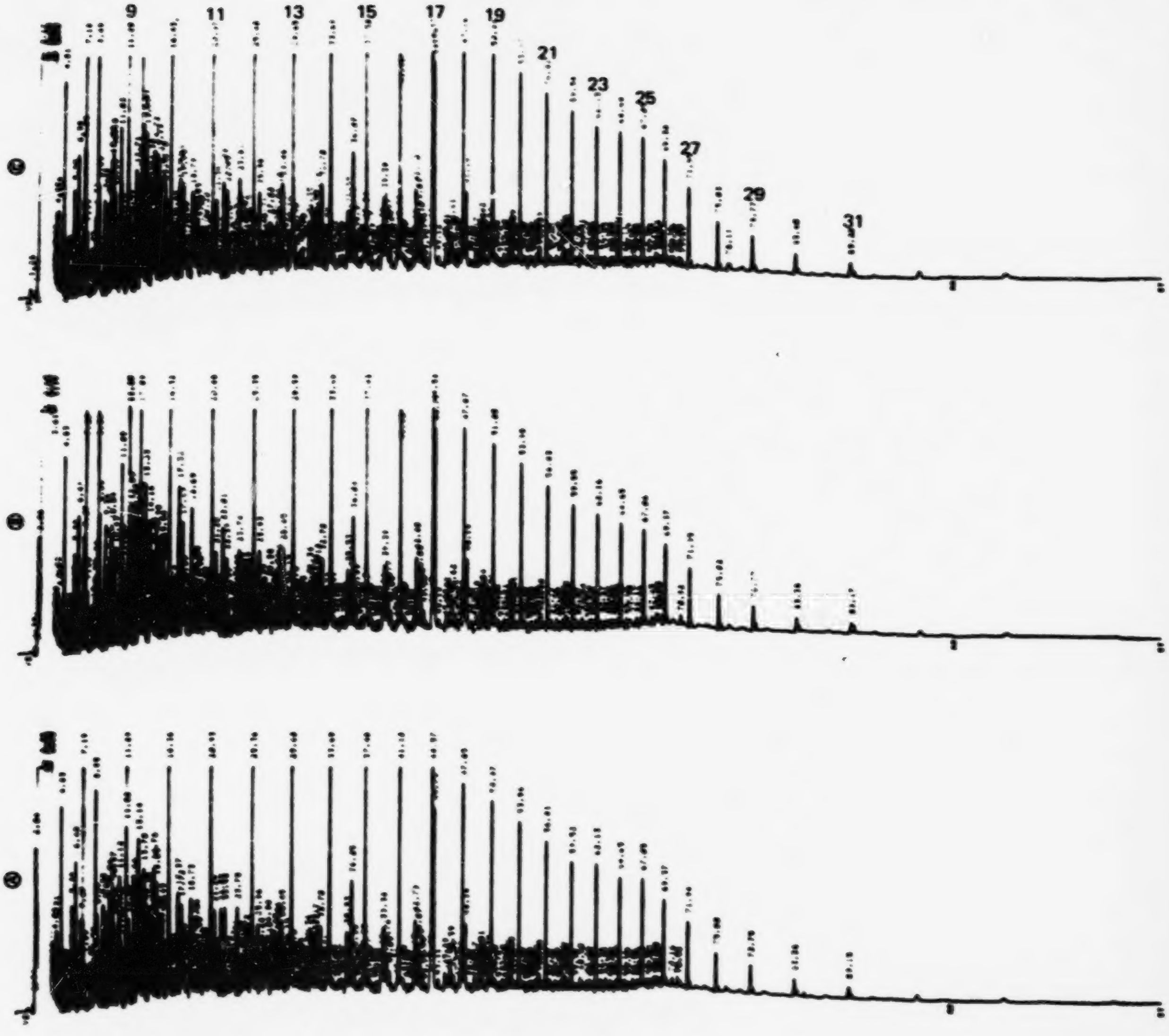

Figure 83. Flame Ionization Detector capillary gas chromatograms of aliphatic fraction extracts obtained on: A, 50 ppt fresh Oil plus starch, B, 50 ppt fresh Oil alone, and C, 50 ppt fresh Oil plus Chitin after one year of natural weathering in the sediments of Sadie Cove.

(c) 50 ppt oil plus chitin samples from Sadie Cove. The three chromatograms are essentially identical showing that little or no degradation of the oil occurred at the 50 ppt level. This is also reflected quantitatively by comparing the numbers in Table 57 for samples Nos. 782, 779 and 780. These data suggest that the total resolved hydrocarbons and unresolved complex mixtures are essentially identical in the three samples. Other similarities include the odd/even hydrocarbon ratios, the ratio of the sum of pristane plus phytane to the total n-alkanes, and the pristane/nC_{17} and phytane/nC_{18} ratios. Essentially, these data suggest that at the 50 ppt level, degradation is not nutrient limited. Figure 84 presents the aromatic fraction chromatograms obtained on the same three Sadie Cove sediment samples: (a) oil plus starch, (b) oil alone and (c) oil plus chitin. As the data in Table 58 illustrate, the aromatic compounds which were identified appear to be essentially the same in all three samples, although there may be some decrease in the levels of aromatic compounds in the oil plus starch sample (a). Replicate analyses would be required to determine if the subtle difference in overall aromatic compound levels is statistically significant. Alternatively, it may be prudent to examine 1.0 ppt oil samples in the presence and absence of nutrients to determine if enhanced aromatic hydrocarbon degradation can be induced to lower overall hydrocarbon levels where the inherent toxicity may be reduced.

IV. Disucssion

A. Implications of hydrocarbon analyses

Many investigators have long suspected that spilled oil on the water surface or in the water column does not consitute as great an

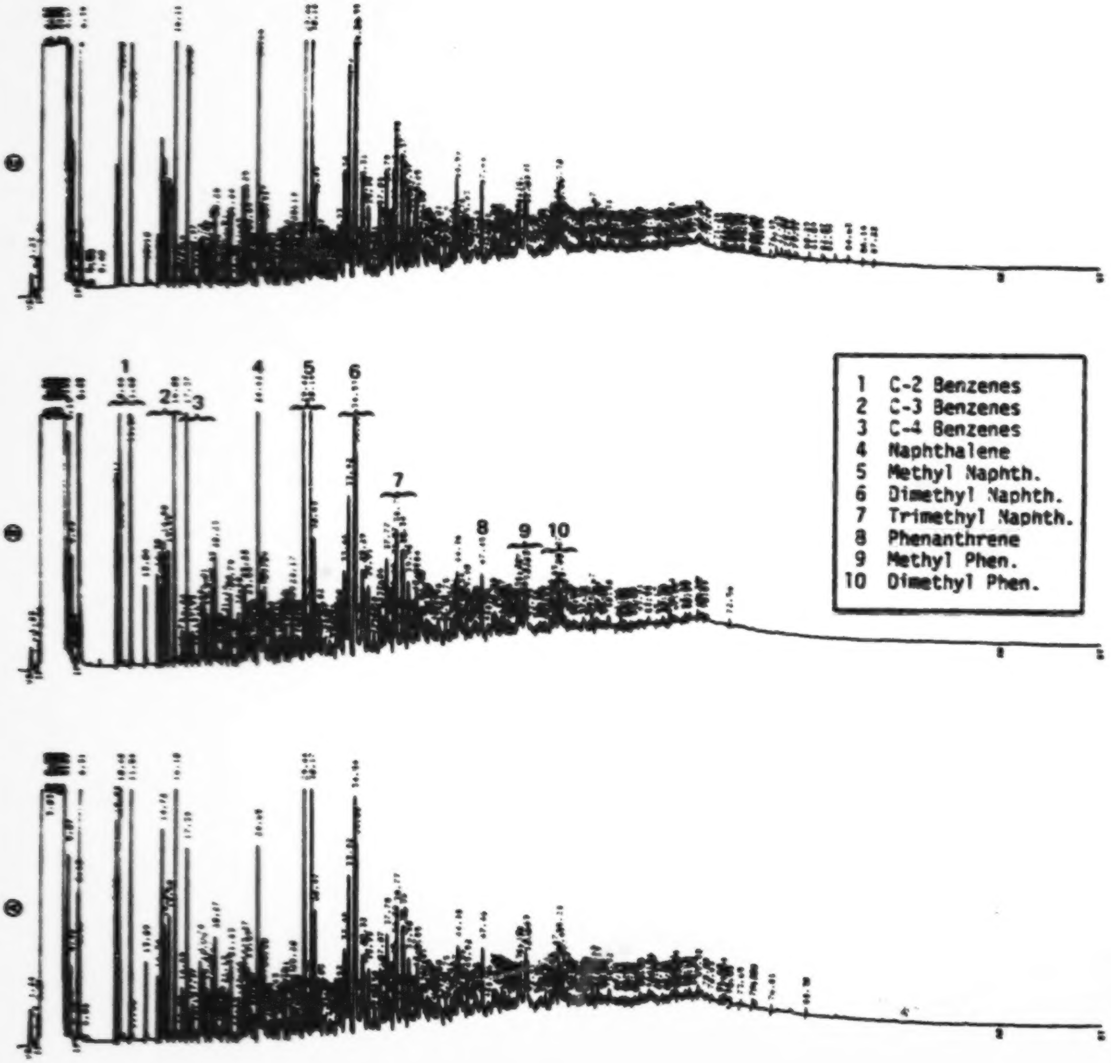

Figure 84. Flame Ionization Detector capillary gas chromatograms of aromatic fraction extracts obtained on: A, 50 ppt fresh Oil plus starch, B, 50 ppt Oil alone, and C, 50 ppt fresh Oil plus Chitin after one year of natural weathering in the sediments of Sadie Cove.

environmental threat as oil which has been incorporated into sedimentary regimes. Ironically, in the case of most major oil spills and laboratory studies, the sediments have been found to be the ultimate repository or sink for the bulk of the higher molecular weight components in the released oil (Jordan and Payne, 1980; D'Oxouville et al., 1979; Meyers, 1978; Mayo et al.; 1978, Gearing et al., 1979; Winters, 1978; Meyers and Quinn 1973; Zurcher and Thuer 1978; Bassin and Ichiye 1977). Once incorporated into the sediments, many of the unweathered toxic components of oil are retained unaltered for extended periods (Teal et al., 1978; Mayo et al., 1978) causing a variety of deleterious effects which are described in detail in Section IV.

If contaminant concentrations reach high enough levels, the biological productivity of an entire area may be completely destroyed immediately after the spill impact, and residual toxic levels may prevent recolonization of native species for a number of years (American Institute of Biological Sciences, 1978). This is a significant problem in areas of high productivity or in sedimentary regimes critical to the survival of juvenile species. Alternately, competing species with different degrees of tolerance to oil could opportunistically recolonize an area, thus further altering the biological balance at the spill site for years.

In this study, we attempted to determine which crude oil concentrations in sub-Arctic sediments would cause long term damage to an area. We also sought to determine concentrations and conditions under which specific compounds in the complex hydrocarbon mixture are selectively removed due to biotic and abiotic processes after incorporation of oil into the sediments. The results of these studies indicate that sediments treated with concentrations approaching

50 parts per thousand (total oil v/v) cause extensive and significant long-term damage to sub-Arctic sediments, and that little or no significant additional weathering (removal of toxic components) occurs at least up to one year following initial exposure. This was observed when both fresh and artificially weathered crude oils were added into the sedimentary matrix at the 50 ppt level. Similar trends were observed at the 1 ppt level, but some evidence of selective lower molecular weight hydrocarbon degradation after one year was found. The experimental results also suggest that a levels of oil approaching 50 ppt, the biotic utilization of specific hydrocarbon components is not inhibited by limited nutrient levels but rather by the toxicity of the oil itself.

B. Interpretation of hydrocarbon data relative to microbial processes

There is substantial evidence in the literature indicating that crude oil degradation requires oxygen, inorganic phosphate and fixed nitrogen and the presence of microorganisms that can degrade hydrocarbons (Colwell and Walker, 1977). In most environments that have been studied, the main limiting factor in crude oil degradation is not the presence of hydrocarbon degrading microorganisms (Atlas, 1977). In pelagic systems, the main limitation to crude oil degradation is usually the absence of sufficient inorganic nutrients. In marine sediments, both inorganic nutrients and oxygen are probably limiting.

If inorganic nutrients were limiting crude oil degradation in the Kasitsna Bay sediments, one would expect to find a depletion of these nutrients in sediments that had been exposed to high concentrations of crude oil. This was not observed in the oil sediments that were analyzed for total phosphate and fixed nitrogen. If oil degradation was limited by only fixed nitrogen, we would expect to observe more degradation in oiled sediments augmented with chitin than those that had not been supplemented with this nitrogen containing compound. The hydrocarbon analysis of the chitin amended sediments indicates that this was not the case. These data suggest that oxygen might have been the main limiting factor in crude oil degradation.

Although we did not routinely measure O_2 levels in the sediments, we did make redox potential determinations in all sediments. In sediments that had been exposed to 50 ppt fresh crude oil, we observed significant reductions in redox potentials within a matter of days after the initial exposure. In sediments that had been exposed to fresh crude oil at 0.1 ppt for one year, we observed a significant reduction (89%) in the redox potential. The hydrocarbon analyses of these sediments showed that there had been essentially total degradation of the original crude oil in the sediments treated at 0.1 ppt, yet there was still a significant reduction in O_2 as indicated by the reduced redox potential.

In both weathered and fresh crude oiled sediments, further reductions in redox potentials were observed when the concentration was increased to 1.0 ppt (Fig. 59). Although there was significant degradation of crude oil in the 1.0 ppt sediments, this was not as complete as that observed in the 0.1 ppt sediments (i.e., a large portion of the aromatic fraction was not degraded in the 1.0 ppt sediments). The hydrocarbon analysis of the sediments treated with 50 ppt fresh crude oil showed that very little, if any, biogenic crude oil degradation had occurred within the first year of exposure. These data suggest that although there may have been direct toxic crude oil effects that inhibited hydrocarbon breakdown, it is also quite likely that O_2 was severely limiting to this process. These data also suggest that the redox potential may remain low in crude oil perturbed sediments after the crude oil has been degraded.

In section IV of this report, we made a number of assumptions which appear correct in view of the hydrocarbon analyses. It was assumed that the sediments that were used for this study had not recently been perturbed by petroleum hydrocarbons prior to the initiation of the study. The hydrocarbon analysis indicate that only biogenic hydrocarbons were present in the nontreated sediments. We also assumed that the "weathered" crude oil used in this study contained very few low molecular weight hydrocarbons. This was also substantiated by these analyses. Essentially all aliphatic hydrocarbons smaller than nC_{13} and the aromatic fraction with molecular weights lower than dimethylnaphthalene were absent in the "weathered" crude oil (Fig. 68).

Another assumption was that the aromatic fraction was the primary cause of the reduced nitrogen fixation rates that were observed. The hydrocarbon analyses suggest that this interpretation is correct. Nitrogen fixation rates were not affected by the presence of weathered crude oil even after being exposed to 50 ppt for 1 year. This means that the inhibition of nitrogen fixation is not caused by the higher molecular weight aliphatic or aromatic hydrocarbons illustrated in Figure 68. In the sediments that had been exposed to 1.0 ppt fresh crude oil for 1 year, there was a very significant reduction in nitrogen fixation rates of 58%. The hydrocarbon analysis of this sample showed that although most of the low molecular weight aliphatic hydrocarbons had been degraded, many of the light aromatic compounds remained.

V. Acknowledgements

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Glossary of Terms

Acute effects -- These are the effects that are observed in samples that had been exposed to the substance being tested for 24 hours or less.

Adenylates -- A group of compounds found in all living things that act as enery transporters within the cell. These include the compounds, ATP, ADP, and AMP.

Amylase -- This is an enzyme that hydrolyzes starch into simple sugars.

Anaerobic fermentation -- This is the degradation of organic nutrients under anoxic conditions. The typical products of anaerobic fermentation are: hydrogen sulfide, methane, ammonia, and organic acids.

Anoxic -- This is a condition where oxygen is absent.

ATP, ADP. AMP -- These are adenylates which are responsible for energy transfer within living cells. Adenosine triphosphate (ATP) contains the most energy, adenosine diphosphate (ADP) contains less energy, and adenosine monophosphate (AMP) is the lowest energy level.

Bacteriovore -- Any organisms that consume bacteria as a food source.

Benthic microorganisms -- Those microorganisms that live in sediments.

Biosynthesis -- This is the process by which new cell material is made.

Challenge experiments -- These are the acute effects experiments where Chitin is one of the major structural components of crab and shrimp exoskeletons.

Challenge experiments -- These are the acute effects experiments where samples are exposed to the substance to be tested for 24 hours or less.

Chitinase -- This is the enzyme that hydrolyses chitin into simple sugars. Chitin is one of the major structural components of crab and shrimp exoskeletons.

Detrital food chain (web) -- This is the food chain which is based on food from detritus rather than from food that is directly consumed as living plant material.

Detritus -- All non-living organic material (including soluble organics) found in the ecosystem.

Direct cell counts -- This is the procedure used to determine the concentration of bacteria by counting them directly under a microscope.

Diversity index -- This is a measure of the number of different types of organisms in a sample.

Epifluorescent microscopy -- This is a procedure that we use to make direct bacterial cell counts. The cells are tagged with a fluorescent stain which can be seen under an ultraviolet light source to distinguish cells from detritus.

Eucaryotic organisms -- Organisms that have cells containing nuclei; i.e., organisms that are higher than the bacteria.

Glucose or glutamic acid uptake studies -- These are studies in which we measure the rate of substrate that is taken up and respired (mineralized) by natural mcirobial populations. The higher the rate, the more active the population. This is often expressed as "relative microbial activity".

Herbivore -- An organism that consumes plants to obtain food.

Heterotrophic potential studies -- Those studies whre we measure the uptake and respiration of organic substrates by natural microbial populations at various substrate concentrations. From these data, we can calculate kinetic variables such as V_{max}, T_t and $K_t + S_n$. Heterotrophic potential = the maximum potential rate at which the test substrate can be taken up and utilized by the microorganisms.

Hydrolase -- Any enzyme that catalyzes a hydrolytic reaction. The hydrolases that we are concerned with (amylase and cellulase) break down large molecules into smaller ones that can be readily utilized by bacteria.

Infauna -- Any organisms that live in marine sediments.

Kinetic data -- Variables that have been calculated from data generated from heterotrophic potential studies.

$K_t + S_n$ -- This is a single value that is estimated during heterotrophic potential studies which includes both the transport constant (K_t) and the natural substrate concentration (S_n); the natural substrate concentration of the same substrate that is added during the experiment. This value can be used in a comparative sense when the S_n value is known not to differ between the samples being compared. Under these conditions, any change in $K_t + S_n$ can be attributed to changes in K_t. If this value increases for some reason, it can then be said that the population will take up the substrate at a lower rate at a given substrate concentration.

Long-term effects -- These are effects that are observed in samples that had been exposed to the substance to be tested for more than 24 hours.

Macrophytes -- A plant that is large enough to be seen by the unaided eye. As used in this context, it means large marine algae that are normally attached to some hard surface.

Mineralization -- This is the process by which organic molecules are converted to inorganic molecules.

Nitrogen fixation -- The process by which atmospheric nitrogen (N_2) is converted to fixed nitrogen; i.e., NH_4, NO_2, NO_3. This is a reaction that requires a great deal of energy.

p value -- This represents the statistical significance of the difference between a set of mean values. A p value of 0.05 means that there is a 95% probability that the difference between the two mean values being compared is not due to chance.

Pelagic microorganisms -- Those microorganisms associated with the water column.

Percent respiration -- This is the percent of the total amount of substrate taken up by the microorganisms that is respired as CO_2. It is calculated by dividing the amount of labeled carbon associated with the CO_2 fraction by tht total amount of substrate taken up by the cells (both cell and CO_2 radioactivity) and multiplying this ratio by 100.

Phosphatase -- This is the enzyme that converts organic phosphate to inorganic phosphate.

Primary productivity -- This is the process by which atmospheric CO_2 is converted to organic carbon. This new organic carbon can be in the form of new plant material or soluble organic material released by the plants during photosynthesis.

Relative microbial activity -- This is a relative estimate of how metabolically active a natural microbial population is. This is determined by the rate at which the microorganisms take up a simple organic compound which is labeled with a radioactive tracer. This is done using two methods; uptake at one substrate concentration or uptake at a series of different substrate concentrations from which kinetic parameters can be calculated. In the multi-concentration method, the kinetic parameter that we use in the determination of relative microbial activity is the maximum potential rate at which the substrate can be taken up (V_{max}).

Secondary productivity -- This is a poorly defined term which generally means the generation of organic nutrients in a form that can be used by animals (not including living plant material). This is a food source that would not be useful to that animal in its original form.

Short-term effects studies -- Effects studies conducted for 24 hours or less.

Suspended particulate matter (also suspended matter) -- This is anything in the water column that will not pass ghrough a membrane filter with a pore size of 0.45 nm.

Transport constant (T_t) -- This is a variable that is calculated from the data generated during a heterotrophic potential determination. This value is the time in hours required for the natural microbial population to utilize the natural substrate concentration of the substrate being tested.

Uptake rates -- Se relative microbial activity.

V_{max} -- This is the kinetic variable calculated from heterotrophic potential determinations which is used as an indicator of relative microbial activity. V_{max} = the maximum potential rate at which the tested population could possibly take up the test substrate.